Scottish Crop Research Institute

Annual Report 1998/99

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The Scottish Crop Research Institute (SCRI) is a major international centre for research on agricultural, horticultural and industrial crops, and on the underlying processes common to all plants. It aims to increase knowledge of the basic biological sciences; to improve crop quality and utilisation by the application of conventional and molecular genetical techniques and novel agronomic practices; and to develop environmentally benign methods of protecting crops from depredations by pests, pathogens and weeds. A broad multidisciplinary approach to research is a special strength of the Institute, and the range of skills available from fundamental studies on genetics and physiology, through agronomy and pathology to glasshouse and field trials is unique within the UK research service.

Das SCRI ist ein führendes internationales Forschungszentrum für Nutzpflanzen im Acker- und Gartenbau sowie in der Industrie und auf dem Gebiet der allen Pflanzen zugrundeliegenden Prozesse. Es hat sich zum Ziel gesetzt, die Grundkenntnisse in den Biowissenschaften zu vertiefen; die Qualität und Nutzung der Kulturpflanzen durch die Anwendung konventioneller und molekular-genetischer Techniken und neuer agrarwissenschaftlicher Praktiken zu verbessern; sowie umweltfreundliche Methoden zum Schutz der Pflanzen gegen Verlust durch Schädlinge, Pathogene und Unkräuter zu entwickeln. Ein breiter multidisziplinärer Forschungsansatz ist eine besondere Stärke des Instituts; und das zur Verfügung stehende Spektrum an fachlichen Ausrichtungen, das von genetischer und physiologischer Grundlagenforschung über Agrarwissenschaften und Pathologie bis zu Gewächshaus- und Feldversuchen reicht, stellt ein einmaliges Forschungsangebot auf den Britischen Inseln dar.

Le SCRI est un centre international majeur de recherche sur les cultures agricoles, horticoles et industrielles et les processus fondamentaux communs à toutes les plantes. Son but est d'accrôitre les connaissances des sciences biologiques fondamentales; d'améliorer la qualité et l'utilisation des cultures par l'utilisation de techniques conventionelles et de génétique moléculaire et par l'application de procédés agronomiques nouveaux; de développer des méthodes de protection moins dommageables pour l'environnement contre les préjudices causés par les ravageurs, les pathogènes et les adventices. L'une des forces majeures de l'institut est une large approche multidisciplinaire de la recherche. L'éventail des techniques disponibles allant des études fondamentales en génétique et physiologie en passant par l'agronomie et la phytopathologie jusqu'aux essais en serres et aux champs est unique au sein du service de recherche du Royaume Uni.

Lo SCRI e' uno dei maggiori centri internazionali nel campo della ricerca sulle colture agricole, orticole e industriali e sui meccanismi fondamentali comuni a tutte le piante. L'Istituto ha come obiettivo principale l'accrescimento del livello di conoscenza delle scienze biologiche fondamentali, il miglioramento della qualità e del potenziale di utilizzo delle colture tramite l'applicazione di tecniche convenzionali o di genetica molecolare e di nuove pratiche agronomiche, lo sviluppo di metodi ecologici di protezione delle colture da agenti patogeni o malerbe. Uno dei punti di forza dell'Istituto e' l'adozione di un approccio largamente multidisciplinare (probabilmente senza eguali nel servizio di ricerca britannico) fondato su una vasta gamma di capacità scientifiche derivanti da ricerche di fisiologia e genetica ma anche di agronomica e fitopatologia supportate da prove di campo o in ambiente controllato.

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Foreword to Annual Report 1998/99

J.E. Godfrey, Chairman of the Governing Body

This has been another remarkably successful year for SCRI. Life science research is developing at a fast rate world-wide and I am very pleased to report that SCRI is at the forefront of plant science research. To maintain our position as one of the world leading research institutes, we have set ourselves some high achievement targets. I am pleased to say we have exceeded these targets. In addition to numerous confidential reports to various sponsors, we have published over 500 papers of which over 300 are refereed. We have 3.3 publications per innovative scientist. Our royalty income and commercial research contracts are rising. Our staff are contributing to many meetings and conferences with several receiving professional accolades and honorary positions. These and many others are achievements to be proud of and I extend my thanks to all the staff who have worked extremely hard.

This annual report will give you an insight into the high quality of the innovative science that takes place at SCRI. I hope you will enjoy reading it.

Introduction by the Director

John R. Hillman

The Scottish Crop Research Institute (SCRI) is a non-profit-making limited company established under the Companies Act, has charitable status and is a Non-Departmental Public Body because over 50% of the total funding is received as grant-in-aid from the Scottish Executive Rural Affairs Department (SERAD), formerly the Scottish Office Agriculture, Environment and Fisheries Department. All members of the Governing Body are appointed by the Secretary of State for Scotland. Staff are not formally civil servants, but are members of the Scottish Office Superannuation Scheme, to become the Scottish Executive Rural Affairs Department Superannuation Scheme 1999. SERAD also funds any redundancies, the site, and much of its fabric and capital equipment. There is also a Management Statement and Financial Memorandum embodying the formal relationship with SERAD. The Pay and Grading System, and Staff and Management Codes are administered by the Biotechnology and Biological Sciences Research Council (BBSRC).

Plants - promoting the creation and protection of wealth and the quality of life.

The Mission of SCRI is:

to sustain excellence and our international reputation for strategic research in crop, plant and related sciences, and to facilitate the application of new knowledge to end-user industries.

The Aims of SCRI are:

* to provide a major international centre for research of the highest quality on agricultural, horticultural and industrial crops important to northern Britain and the rest of the World, by sustaining a broad, yet fully integrated programme of fundamental, strategic and applicable research designed to contribute to, and complement other sectors of the UK science base;
* to increase fundamental knowledge in the biological sciences while improving crop quality, utility and value through the application of conventional and novel molecular genetic breeding techniques and improved agronomic practices, and by developing more sustainable, environmentally sensitive methods to protect crops from depredations by pests, pathogens and weeds;
* to create wealth and protect investment in our essential plant-based industries by exploiting the advantages and solving the problems of crop production in northern Britain while seeking to improve the quality of life and safeguard the global environment;
* to promote public awareness and understanding of relevant environmental and bioscience issues through technical and lay publications and targeted presentations;
* to encourage, train and reward staff with relevant skills in crop genetics, plant biotechnology and physiology, chemistry, plant pathology, biomathematics and environmental studies, agronomy and the field trialling of new crop varieties.

SCRI was established in 1981 by an amalgamation of the Scottish Horticultural Research Institute (SHRI, founded at Invergowrie, Dundee in 1951) and the Scottish Plant Breeding Station (SPBS, founded at East Craigs, Edinburgh in 1921). In 1987, the Institute assumed managerial responsibility for the Biomathematics & Statistics Scotland (BioSS), formerly the Scottish Agricultural Statistics Service.

SCRI is a major international centre for basic, strategic and applied research on agricultural, horticultural and industrial crops and on the underlying biological processes common to all plants. It is the only such Institute in Scotland, and the range of complementary skills assembled at the Institute, from fundamental molecular genetics to glasshouse- and field-trials, with exploitation of the SCRI-based international genetic resources in a region of high phytosanitary conditions is unique within the UK.

The science is optimised by a matrix management system comprising (w.e.f. 1-10-99) four divisions and nine inter-related research units. Management structures are regularly reviewed to ensure maximum effectiveness of the research at SCRI.

In the past year the Institute has initiated a major Science Strategy Review. The review team will:

- Assess the quality, novelty, focus and timeliness of current and projected research objectives and their relevance to SCRI's position within Scottish, UK and International plant science communities.
- Examine the management of science within the Institute, including the effectiveness of communication and synergies within and between Units and Departments.
- Address the issues raised by the recent Visiting Group and identified in the (1999) SOAEFD Strategy Review.
- In the light of the above make recommendations on the future research direction and integration of research activities. Wealth creating potential and industrial relevance are considered as high priority.

The review will take account of discussions with staff, senior management, the Governing Body and SERAD.

The SCRI research programmes are peer reviewed at many levels. Each year the 'core' programme of research comprising a number of projects is assessed by the Research, Education and Advisory Services (REA) Unit of SERAD. New projects are appraised by advisers prior to commissioning, progress is monitored annually and ultimately a final report is produced for evaluation.

Every 4 years SERAD commission BBSRC to appoint a Visiting Group to review the work of the Institute. A Visiting Group, initially scheduled for June 1998, took place in November 1998. The Group comprised:

Dr C A Gilligan (Chairman)
University of Cambridge

Professor K R Barker
North Carolina State University, Raleigh, USA

Professor M J Daniels
Sainsbury Laboratory, Norwich

Professor J Draper
University of Wales, Aberystwyth

Professor M J Emes
University of Manchester

Professor R W Goldbach
Wageningen Agricultural University, The Netherlands

Professor G J Jellis
Plant Breeding International Cambridge

Professor M J Kearsey
University of Birmingham

Professor A G O'Donnell
University of Newcastle-upon-Tyne

Dr R M J Storey
British Potato Council, Oxford

Dr R G Turner
British Society of Plant Breeders Ltd, Ely

Mr J H Wyllie
Ruchlaw Produce Company Ltd, Dunbar

The Visiting Group's secretariat was provided by Mr B Harris of BBSRC, assisted by Mr J B Sykes and Miss C M Dow. The Group was also accompanied by Dr A J Rushworth, Dr R N Waterhouse and Dr T W Willison of SERAD's Research, Education and Advisory Services Division.

The task of the Visiting Group was: (a) to carry out a scientific audit of the quality and conduct of SCRI's core research programme and related work; and (b) to assess SCRI's effectiveness in managing its resources to meet the needs of users and beneficiaries of research, and the Institute's strategic plans for the future development of its scientific programme.

Remarks published in the Report of the Visiting Group include:

- The VG was impressed by the quality of research at SCRI
- Overall, SCRI's productivity was similarly impressive
- The calibre of SCRI's staff was considered to be good and the VG identified several outstanding individuals.

The Report made a number of recommendations to guide the continuing development of the Institute. A response to the Report has been forwarded to SERAD and an implementation programme is being prepared.

A broad multidisciplinary approach to fundamental and strategic research, and technology transfer are unique strengths of SCRI. Our programmes span the disciplines of genetics and breeding, molecular and cellular biology, biotechnology, plant pathology (bacteriology, entomology, mycology, nematology and virology), plant physiology and cell biology, environmental science, plant chemistry and biochemistry, agronomy, molecular ecology, vegetation dynamics, bioremediation, serology, physics, mathematics, bioinformatics and statistics. Genetics and enhanced breeding of selected crops, and biotechnology lie at the core of all our substantial research, development and training programmes.

The breadth and depth of knowledge, technical expertise and infrastructural resources available at SCRI attract extensive contracts and consultancies from, and foster collaborations with, numerous academic and corporate organisations around the World. Synergistic liaisons with other institutes, universities and colleges in the UK and overseas are also integral to the scientific growth, development and validation of the Institute's research activities. New links are being forged continuously, as well as existing contacts being developed and strengthened.

SCRI and Mylnefield Research Services (MRS) Ltd, the commercial arm of the Institute, are successful in gaining competitive research contracts from government departments and agencies, Levy Boards, grower organisations, international agencies, the European Union, commercial companies, local government, and some Charities, Research Councils and Trust funds, although we are largely excluded from submitting applications to the latter three sources.

SCRI provides the base and secretariat for The Scottish Society for Crop Research (SSCR), a registered Friendly Society formed in 1981 by the amalgamation of The Scottish Society for Research in Plant Breeding and The Scottish Horticultural Research Association.

The SSCR provides an important link between SCRI research scientists and farmers, growers, processors and other interested companies in the private sector.

The Society:

- organises interactive field walks and end-user/researcher discussion sessions;
- finances science-based advisory publications for the benefit of its members;
- stimulates crop-based sub-committees to support targeted research projects ;
- reinforces SCRI representation with trade associations, Levy Boards, and other user-groups;
- administers the biennial Peter Massalski Prize to the most promising young scientist at SCRI.

SCRI is one of five Scottish Agricultural and Biological Research Institutes (SABRIs; Scottish Crop Research Institute, Hannah Research Institute, Macaulay Land Use Research Institute, Moredun Research Institute, Rowett Research Institute) and together with the Royal Botanic Garden, Edinburgh, the Scottish Agricultural College (SAC), the Scottish Agricultural Science Agency (SASA), the Fisheries Research Services and Forestry Commission Research Agency, comprise the Committee of Heads of Agricultural and Biological Organisations in Scotland (CHABOS).

BioSS was established to cover the biomathematical and statistical needs of the five SABRIs and SAC. High-level consultancy, training and research inputs from BioSS give a major advantage to the SABRI and SAC research programmes, as well as to the work of SASA and several other bodies for whom it carries out contracts.

This Report details only a small selection of the recent research achievements of SCRI, BioSS and MRS Ltd, briefly describes the commercial rôles and successes of MRS Ltd; and summarises the important linking rôle of SSCR. Significant advances continue to be made in both fundamental and strategic science, with contributions to the protection and understanding of the environment. In the past year SCRI has contributed to the debate on genetically modified crops, providing independent and unbiased information on this important subject. Discoveries are reported of direct and indirect benefit to agriculture, horticulture, forestry, land management and biotechnology. Dedicated and talented scientific and support staff in the Institute, and BioSS, and MRS Ltd., account for our stature, successes and delivery of achievements.

Details of the annual accounts, Corporate Plan, health and safety provisions, and the

SCRI/MRS quality assurance arrangements are available on request.

On behalf of the staff and Governing Body, it is a pleasure once again for me to acknowledge with gratitude the staff of SERAD for their continuing support of, and demonstrable commitment to, our research programme and to our development. Regardless of the enormous pressures upon them in recent years, they function rigorously, openly and fairly, as always, to the highest professional standards of British public service. Grants, contracts, donations, advice and joint participation in our activities from the SSCR, other government departments and their agencies, non-governmental agencies, our sister CHABOS institutions and BBSRC institutes with whom we coordinate our research, grower levy boards, local and regional authorities, commercial companies, farmers and other individuals, and learned societies, are also warmly appreciated.

SCRI remains buoyant in generally difficult times for science in the UK, justifying its existence in every respect. We have every confidence in meeting future challenges. Scientifically and commercially, our prospects are outstanding.

Report of the Director

John R. Hillman

Global perspectives of factors influencing agricultural, biological and environmental sciences, and their associated industries : 1998-1999*

Preamble

As the new century heralds the new millennium, the historical records will bear testament to a remarkable 20th century, one of scientific, engineering and technological achievement. At the fin de siècle, discoveries, inventions and concepts relating to the biological and environmental sciences were of primary importance to the development of civilisation, offering stunning opportunities for scholarship and wealth creation, but causing concern to those unversed in, or antagonistic to, matters scientific or technological. The influence on the public and politicians of pressure groups or special interest groups opposed to scientific and technological advancement, capitalism, globalisation, multinational corporations, profitability, or even aspects of current agricultural and medical practice, was aided in Europe by compliant broadcast and publishing media. Many non-scientists sought cognitive status for the arts and humanities, accusing scientific realism of producing modern-day technocratic, bureaucratic and relativistic tyranny; science without epistemological, ethical and ideological awareness was claimed to worsen problems for humanity. There was a general and blatant lack of appreciation of the difference between the activity of science – the pursuit of truth and understanding - and its use, which can sometimes raise ethical issues, and sometimes discomforting truths and choices; almost invariably, though, science provides the forward momentum for improvement of the human condition.

The difficulties faced by scientists in the public sector in Europe did not ease during 1998-1999, even though private-sector and charitable support of research and development (R&D) continued to increase, particularly in those areas of the life sciences relating to medicine and the environment. The agricultural biotechnology (agbiotech) sector received massive support outside the UK. For UK public-sector scientists in general, there were continuing severe budgetary constraints, a decline in status in the eyes of the public following regulatory failures

* This review updates and enlarges on themes developed in my previous accounts in the *SCRI Annual Report* series.

and a poor image in the media, and greater bureaucratic interference.

Global output and the volume of world trade in goods and services grew at a slower rate than the year before, and commodity prices fell sharply. There was a slight decline in world agricultural production. By the end of 1998, the impact of the Asian economic crisis was felt in most of the world's economies.

Scientific progress was manifest in diverse areas of scholarship during 1998-1999. The following represent but a few examples of advancements in knowledge presented in the literature. As a result of correlations between the geological record of plant and animal fossils and the geochemistry of minerals, it was possible to date the ages of rocks with greater accuracy than hitherto. S A Bowring and D H Erwin in the USA used high-resolution uranium-lead geochronology to determine zircon ages with uncertainties of less than one million years. They concluded that the age of the beginning of the Cambrian Period was 543 million years, considerably younger than had been considered before. Such precision assisted in calculating the rates of evolution of species. The Cambrian explosion of life, a geological layer associated with the appearance of a wide range of fossils, was faster than previously recognised, lasting no more than 10 million years. Using high-precision mass spectrometry, there was evidence that the mass extinction of life forms at the end of the Paleozoic Era (now dated at 251 million years ago), at a time when 85% of all marine species, about 70% of land vertebrates, and many plants and insects disappeared, apparently occurred in less than one million years. The cause of the extinction is unresolved but is scientifically fascinating.

Paleontological studies provided indirect evidence that angiosperms, the flowering plants, existed during or even prior to the late Jurassic Period (163 million to 144 million years ago). Fossil angiosperms of the Early Cretaceous Period (144 million to 97.5 million years ago) are well described. D Ren of the National Geological Museum of China discovered fossil orthorrhaphous Brachycera flies in late Jurassic rocks. The flies had mouthparts and body hairs characteristic of extant members of the group that are mostly nectar feeders and pollinators.

In mathematics, T Hales of the University of Michigan announced a proof relying on computer verification of the face-centred cubic packing conjecture of Johannes Kepler, the German mathematician and astronomer, in 1611. The conjecture was originally stimulated by Walter Raleigh who sought a rapid way to determine the number of cannonballs in a pile with a base of any shape. Kepler concluded that the manner in which greengrocers commonly stack round fruit such as oranges – a square-based pyramid with each layer of oranges sitting in a square grid centred above the holes in the layer below – gives the densest or tightest way to pack spheres in infinite space. In 1831, the German mathematician C F Gauss showed that the orange-packing design would not be less dense than other lattice packings. Certain non-lattice packings, however, are almost as efficient and may be superior for dimensions higher than three.

In physical chemistry, scanning tunnelling microscopy demonstrated single-molecule rotors revolving inside bearings of other like molecules. R C Cauble and associates in the Lawrence Livermore National Laboratory, California, and the University of British Columbia, reported the first experimental evidence for the transition of hydrogen to a metal with superconducting properties. Deuterium, an isotope of hydrogen, was chosen to compress to 300 GPa using a laser beam because it is easier to compress than hydrogen, but hydrogen would be expected to behave in the same way. This raises the possibility that the giant gas planets (*e.g.* Jupiter and Saturn) have gravitationally induced metallic hydrogen cores.

In 1998, physicists using the Super-Kamiokande facility in a zinc mine near the village of Kamioka, Japan, provided the strongest evidence yet that the subatomic neutrino particle, specifically the muon neutrino, has detectable rest mass. The standard model of the fundamental constituents of the universe involves three families of particles: baryons (*e.g.* neutrons and protons); leptons (*e.g.* electrons and neutrinos) and mesons. Neutrinos interact so weakly with other matter that they are difficult to observe, and were therefore assumed in the model to lack mass. Three types or flavours of neutrinos were known, each closely associated with the production of its charged lepton namesake: electron neutrinos emitted in nuclear beta decay when electrons are emitted; muon neutrinos derived from the decay of cosmic-ray-produced pion particles, and tau neutrinos produced from tau particles, the latter particle having been observed for the first time in 1998 at the Fermi National Accelerator Laboratory. That neutrinos have a mass will cause a revision of the standard model, and may well explain the deficiency of current cosmological models of the

universe which require it to have a mass far in excess of the total mass of readily observable constituents.

Einstein's general theory of relativity was reinforced by astronomers from France, The Netherlands, UK and USA. Using an infra-red camera on the Earth-orbiting Hubble Space Telescope, they exploited gravitational lensing, in which light from a distant object is focused as a result of the distortion of space by a massive foreground object such as a galaxy. They observed an Einstein ring in which the image of the light source formed a perfect ring around the foreground object. By the end of 1998, there was evidence of the existence of 22 planets outside our solar system, most with orbital periods of only a few days, and highly elliptical orbits – far from promising targets for astrobiology. R H Mendez and colleagues at the Munich University Observatory, H Ferguson of the Space Telescope Science Institute, Baltimore, and colleagues using the Hubble telescope, provided evidence of the existence of substantial populations of isolated stars in intergalactic space.

For the first time for a complete multicellular animal, teams from the UK and USA working on the Human Genome Project, sequenced the entire genome (six chromosomes comprising 97 million base pairs giving rise to about 20,000 protein-coding genes) of the 959-celled nematode *Caenorhabditis elegans*. The genomes were also sequenced of *Chlamydia trachomatis*, *Mycobacterium tuberculosis*, and *Treponema pallidum*. In October 1998, 64 scientists produced a new human gene map (GeneMap '98) marking the chromosomal locations of more than 30,000 genes (http://www.ncbi.nlm.nih.gov/genemap/). A working draft of the three billion genetic letters of the human genome is expected to be produced by the spring of 2000. Current estimates put the total number of genes at around 100,000-140,000 and the next major challenge will be to unravel how the genes function. Advances were made in understanding the rôle of telomerase in cellular senescence and cancer, and in the possible use of endostatin and angiostatin in controlling angiogenesis - localised blood vessel formation to supply malignant tissues. Other major advances were made in stimulating the regeneration of neurons and in unravelling the rôle of interleukin-13 in asthma. Hope was offered for the control of diseases such as sleeping sickness and Chagas' disease caused by parasitic protozoans; hypoxanthine/guanine phosphoribosyl transferase (H/GPRTase), used by the protozoan to salvage purines from the host, can now be discriminated from the host H/GPRTase, offering a target for curative treatments. Antibiotic-resistant strains of *Mycobacterium tuberculosis*, *Salmonella typhimurium* and *Streptococcus pneumoniae* were recorded in both Less-Developed (LDCs) and More-Developed Countries (MDCs). Viagra (sildenafil), the first oral drug for male impotence, was released in a blaze of publicity. J Ma of Guy's Hospital, London, created a caries vaccine in tobacco plants by transferring into the plant genes for antibodies against the major cause of caries, *Streptococcus mutans*. Fresh fruit and vegetables will be logical market targets for the technology.

Referred to variously in the media as the 'millennium bug', the 'millennium bomb', the 'year 2000 problem', the 'Y2K bug', or the 'Y2K problem', it is likely that difficulties may arise through computer programming shortcuts taken mainly in the 1970s and 1980s. This predicament has arisen by (i) abbreviating four-digit years to two-digit years in order to save memory space, (ii) sometimes failing to programme in the year 2000 as a leap year, and (iii) not taking into account the fact that some computers may record 9 September 1999 – *i.e.* 9/9/99, a series of nines – as the end of a program. The Y2K problem is not restricted to computers but also to devices containing computer chips, *i.e.* embedded systems. Government services, public utilities, financial services, transport, communication systems, emergency services, defence systems, hospitals, research organisations *etc*. were all thought to be vulnerable. In December 1998, the United Nations (UN) convened its first international conference on the Y2K problem, which according to the Gartner Group could cost as much as $300-$600 billion to correct.

Estimates by Forrester Research illustrated the rapid development of Internet retailing. In 1998, consumers in the USA were thought to have purchased $7.3 billion of goods over the Internet, double the 1997 total, and on-line sales were expected to increase by an additional 65% in 1999 to about $12 billion. Internet retailing appealed strongly to investors in the so-called 'dot.com' companies despite intense price competition and low or, more usually, negative profit margins. Governments have yet to agree on encryption (encoding) software to facilitate electronic commerce, and have yet to come to terms with the taxation implications of e-commerce and cross-border 'mobile activity', and currency blocs could become less important.

International aspects of education common to many countries concerned achievement testing at the pri-

mary and secondary level, expansion of information technology as a teaching aid, trans-national co-operation in higher education, and financing and quality checks of schools and universities. Some countries experienced student protests (*e.g.* Brazil, India, Russia, Serbia) and violence (*e.g.* USA). There were controversial issues over the schooling of girls and young women, and the teaching of the Qur'an (Koran) in some predominantly Muslim countries, and the teaching of evolution in the USA. In higher education, the era of the electronic or virtual university was beginning to exercise strategic planners. International co-operation was a pronounced feature in 1998, as evidenced in greater recognition of the European Union (EU) Erasmus programme for the exchange of students between EU nations. Many countries devised systems to attract able foreign students, by offering attractive tuition regimes, grants, distance tuition, new courses and institutional cross-links. Most publicly funded higher educational institutions in the MDCs and the LDCs were forced to operate with diminishing financial resources in an internationally competitive market place ripe for the full incorporation of information technology and new teaching methods.

One particular area of concern was the growing menace of 'junk' science to corporations as well as insurance and other companies in the civil justice system. This hotch-potch of speculative theories, poor statistics, and questionable honesty or independence of expert witnesses, was aided by the apparent faith of the public in the incorruptibility of its proponents. The reader is recommended to consult *Science on Trial* by M Angell.

It was appropriate that 1998, the year that marked the 50^{th} anniversary of the Universal Declaration of Human Rights, should also be noteworthy for the establishment by treaty of the International Criminal Court. Of the 148 countries involved in discussing the setting up of the court, 120 voted in favour, 21 abstained and seven (including the USA) voted against. Crimes to be covered by the treaty included genocide, crimes against humanity and war crimes, but regretfully not terrorism nor drug trafficking. Official ratification of the treaty will require signatories of at least 60 nations, a process that could take several years, notwithstanding the declared intention of support from double that number of nations. To many, the most serious limitation in the court's jurisdiction was that it could act only when the nation, territory or nationality of the accused has become party to the treaty or had consented to do so. Two fundamental flaws still remain at the outset, one of claiming jurisdiction over the objecting nations (principally the USA and China), and two, diluting the authority of the UN Security Council.

At the end of the year, the US House of Representatives approved two articles of impeachment against the President of the USA, W J Clinton, propelling him towards a damaging but inconclusive Senate trial in the first part of 1999. The media-feeding frenzy on his private pastimes did not seemingly depress his high approval rating among voters.

Financial collapse loomed over the UN in 1998, exacerbated by the failure of the USA to pay its full dues to the 185-member inter-governmental organisation, a situation that has prevailed since 1995. By the end of 1998, the USA owed $1.8 billion. This deficit was temporarily offset by (i) the provision of interest-free loans, some from LDCs; (ii) contributions by non-governmental organisations (NGOs); (iii) failure to repay countries for providing peace-keeping troops; and (iv) additional economies imposed on the 15 independent specialised agencies (*e.g.* the Food and Agriculture Organization of the United Nations - FAO, International Fund for Agricultural Development, World Health Organization - WHO). Serious political challenges to the UN came from the behaviour of Iraq, the former Yugoslavia, the Democratic Republic of the Congo, and the countries in the Horn of Africa. Iraq abruptly terminated the work of the UN Special Commission (UNSCOM) charged with destroying its weapons of mass destruction. NATO and the UN tried to address atrocities in Kosovo, and the UN was forced to suspend investigations of massacres in the Democratic Republic of the Congo.

For the most part, the attention of the Commonwealth of Nations was directed towards West Africa, to foster and promote the development of democracy in Sierra Leone and Nigeria. Commonwealth countries also led moves to restructure and force international financial institutions to provide debt relief for poor, largely agrarian countries. The Commonwealth is a multi-racial voluntary association of 54 Sovereign independent states; with the exception of Mozambique, all were formerly parts of the British Empire or League-of-Nations mandated territories. Agriculture is an important inter-governmental link, and assistance to other Commonwealth countries normally has priority in

bilateral aid programmes of the association's four MDCs (Australia, Canada, New Zealand, and UK). In January 1998, the UK government announced that 13 of the remaining UK-dependent territories would be re-categorised as British Overseas Territories.

In the first half of 1998, India and Pakistan conducted, to international displeasure, a total of 11 nuclear tests and joined the five acknowledged nations possessing nuclear weapons. Iraq was attacked by aircraft from the USA and UK at the end of the year for failing to comply fully with the UN inspectors. War in the Balkans spread inexorably to the Serbian province of Kosovo. The civil war in the Democratic Republic of the Congo (the former Zaire) continued remorselessly and involved troops from Angola, Chad, Namibia and Zimbabwe on the side of President L Kabila, facing troops from Rwanda and Uganda supporting the rebels. A UN report estimated that about 300,000 children under the age of 18 were serving world-wide as combatants.

In respect of arms control, Brazil signed the Nuclear Non-Proliferation Treaty, leaving Cuba, India, Israel, and Pakistan as the only nations that had not signed. France and the UK were the first nuclear powers to ratify the comprehensive Test Ban Treaty, which needs to be ratified by the 44 nuclear or potential nuclear states to come into effect.

Towards the end of 1998, 133 nations had signed the 1997 Ottawa Convention banning the use, stockpiling, production and transfer of antipersonnel land mines, and 59 nations had ratified it. The USA maintained that such mines were needed to defend the demarcation line between North and South Korea. Also, by the beginning of 1999, 169 nations had signed or acceded to the Chemical Weapons Convention, prohibiting the development, production, possession or use of chemical weapons, and mandating the destruction of stockpiles by 2008. Financial problems may prevent Russia from meeting the deadline to destroy its 40,000 tonnes of chemical-weapon agents.

In NATO, by the end of 1998, all of the 16 members except The Netherlands had ratified the accession protocols for bringing the Czech Republic, Hungary, and Poland into the alliance. The three new members were inducted formally in April 1999. Relations with Russia became distinctly frosty and edgy over disagreements with NATO policy on dealing with conflicts in the Balkans, and Russia threatened to abrogate the 1997 Founding Act regulating its special relationship with NATO. Preparations were put in hand by NATO to launch a bombing campaign against Yugoslavia (Serbia) following the inability of the Security and Co-operation in Europe observer force to verify that Serbian forces had been withdrawn from Kosovo.

Terrorists launched co-ordinated attacks in August on the embassies of the USA in Kenya and Tanzania, precipitating counter-terrorist attacks by the USA in Afghanistan and The Sudan. The fragile peace accord in Northern Ireland was strained by the August bombing incident in Omagh, killing 28 people and injuring 220. It was the worst atrocity in Northern Ireland in almost three decades, and was claimed by the Real Irish Republican Army. In September, the Basque terrorist organisation, Euskadi Ta Askatasuna (ETA) announced an indefinite cease-fire.

Economics and Politics

By mid-1998, the impact of the Asian economic crisis began to affect other economies, starting with the declaration of a debt moratorium by Russia. In August 1998, Russia devalued its already weakened currency, defaulted on a large portion of government debt, and stopped foreign credit repayments by companies and banks. Instability and uncertainty affected confidence in the bond markets and emerging economies. There was a real possibility voiced that the western industrial economies, having failed to avoid a downturn in the financial markets, might become embroiled in a recession. Certainly, the Japanese economy with its heavily indebted banks was far more severely affected than first thought, indicating the start of a deflationary economic environment where there was a weakening demand for goods. By the end of 1998 and the first part of 1999, economic forecasts were being downgraded, indicating proximity to recession. Some worried commentators likened the serious financial crisis to the Great Depression of the 1930s.

In 1998, the economies of Indonesia, South Korea, and Thailand were expected by the International Monetary Fund (IMF, see later) to shrink by 15%, 7% and 8%, respectively. Japan's economy had moved into its worst recession since 1945 with its Gross Domestic Product (GDP) falling by 2.8% in 1998. Economic growth in China, much of Latin America, and Africa also slowed. Parenthetically, the GDP is defined as the total value of the final goods and services produced within a country during a financial or calendar year. The System of National

Accounts 1993 – published under the auspices of the UN, IMF, Organization for Economic Co-operation and Development (OECD), EU and World Bank – provides the universally accepted framework for international comparability in classifying and presenting domestic accounting aggregates and international transactions comprising 'net factor income from abroad', the measure that distinguishes GDP and Gross National Product (GNP). Thus, GNP is the total value of final goods and services produced both from within a given country and from foreign (external) transactions in a financial or calendar year. The net factor income from abroad is defined as the income residents receive from abroad for factor services (labour, investment, and interest) less similar payments made to non-residents who contribute to the domestic economy. For the UK, as in all member states of the EU, the national accounts are being updated to new methodologies given in the European System of National Accounts 1995. The UK moved on to the new basis of national accounts with the 1998 Blue Book published by the Office for National Statistics. A common methodology was agreed in 1997 (*Manual on Economic Accounts for Agriculture and Forestry* produced by Eurostat) for aggregate agricultural accounts for all member states of the EU.

Hitherto, the Asian economies had been fêted as paradigms of economic virtue and growth, but closer analysis reveals that Asia's rapid growth was attributable to high savings and investment rates, unjustifiable confidence in future prosperity, and relaxation of central capital controls restricting foreign investments (bank loans, portfolio investments, bond purchases and direct investments into existing and new companies). Between 1990 and 1996, Indonesia, Malaysia, Philippines, South Korea and Thailand received about $300 billion in foreign investment, providing the classic conditions for a 'boom' as a prelude to a 'bust'.

Although the specific origins of Asian crisis differed from country to country, there was a commonality in unsound financial and banking systems, overvalued assets, and political interference in commercial investment decisions. Much of the capital inflow had not generated adequate returns, even to repay interest and principal. The economic deviation quelled importation demand, affecting Western exporters, unemployment rose rapidly, and there was a slump in confidence in the Asian economies. Most of the Asian countries in crisis had received loans from the IMF and other international agencies in return for commitments to improve bank, market and project regulation. A 1998 report from the UN Conference on Trade and Development in 1998 criticised the Big Five accounting firms for giving a clean bill of health to many large Asian companies and banks, despite their flouting of international accounting standards. The report stated that fund managers, as well as analysis, and credit-rating agencies needed also to accept blame for allowing huge influxes of money into potentially weak enterprises in the region.

It was the collapse of the Thai baht in July 1997 that triggered the Asian economic crisis, causing uncertainty for the world economy. The compounding effects of the recession in Japan, the repercussions of the financial crisis in East and Southeast Asian countries, and investor behaviour led to the IMF revising down its projections for world economic growth from 4% to 2%. All emerging markets were adversely affected, provoked further by the August financial collapse in Russia, such that by September 1998 the financial turbulence spread to varying extents to the 29 developed countries of the OECD. Measures to address the turbulence included reductions in interest rates, falling stock-market prices, new legislation or processes to recapture investor confidence by reforms of the banking system, and support loans. Currency devaluations frequently associated with declining commodity prices had severe effects, not least on agricultural commodity trading, and hence depressing or negating the profitability of agriculture.

During 1998, output in the major industrialised countries rose by an average of 2%, compared with 3.1% in 1997. Some countries suffered sharp deterioration of their output figures, however, including Japan, South Korea, Taiwan, Hong Kong, and Singapore. By way of contrast, the economy of the USA was resilient in its buoyancy, with output rising by 3.5%. Likewise, increases were posted in the EU where output rose on average to 2.9% compared with 2.7% in 1997. Both the USA and the EU benefited from strong domestic demand. Output in Central and Eastern Europe rose to 3.4%, but only Poland, Slovakia and Slovenia regained their 1989 levels of output. A 6% decline in Russia led to a slight overall decline in output of the former centrally planned economies.

The growth rate of output in the LDCs fell from 8% in 1997 to 2.3% in 1998. On a *per capita* basis, GDP grew by 0.7%, a marked fall from the previous 6 years in which GDP *per capita* grew by 4% or more. The

latest output projection for 1999 was for 3.6%, dependent on commodity-price movements. A 3.7% rise in the GDP of Africa in 1998 reflected financial restructuring and beneficial weather for agriculture in some countries, such as Algeria and Morocco. No output expansion was expected in South Africa. Reduced outputs in the Middle East (2.3%) and in Latin America (2.8%) were associated with sharply falling oil prices. Output in Asia was constrained for the first time in two decades to 1.8% by the economic crisis, although China and Taiwan were apparently more resilient than most.

The Association of Southeast Asian Nations (ASEAN), which includes Brunei, Indonesia, Laos, Malaysia, Myanmar (Burma), the Philippines, Singapore, Thailand and Vietnam, was sorely tested by the Asian financial and economic crisis. Overt reluctance of the ASEAN foreign ministers at their meeting in July 1998 to analyse the internal affairs of members was widely seen as a set-back to the move towards an ASEAN free-trade area by 2003. Mercosur, the Southern Cone Common Market, reported in 1998 that since its creation in 1991, the total international trade of its member countries had doubled, and it had become the fourth largest trading bloc in the world. The possibility of a common currency in the region by 2010 was discussed during the year, emulating the processes taking place in the EU. In 1997, the last year for which reliable figures are available, around 55% of the total aid to Latin America came from the EU. In trade, however, according to the Madrid-based Institute for European-Latin American Relations, the value of EU trade with Latin America was less than that of its trade with Switzerland. This was mainly because the CAP limits the sale of Latin American agricultural goods by virtue of tariff protection and export subsidies. A full free-trade agreement would need a seismic shift in the CAP and the attitudes of those countries which are net beneficiaries of CAP-based assistance.

In terms of volume, international trade of goods and services rose by 3.7%, decelerating rapidly from the 9.7% level of 1997 which had marked a new annual peak in international trade. In value terms, the $6.6 billion trade in 1998 was similar to that in 1997, reflecting the dramatic fall in commodity prices and the weakening of prices of manufactured goods. LDCs suffered major declines in imports and exports. For the first time, the group of four newly industrialising countries (Hong Kong, Singapore, South Korea and Taiwan) were a negative source of trade momentum, whereas, for most of the last three decades, these countries enjoyed double-digit export and import growth. Widespread acceptance of the need to open up markets was evident by the agreement of 70 members of the World Trade Organization (WTO), representing 95% of global markets, to liberalise financial services further. The commitment to free up markets was given backbone by governments rejecting protectionism at the WTO meeting in May 1998.

The next round of trade liberalisation talks at the WTO is scheduled for November 1999, and will include the 'built-in' agenda agreed in the 1986-1993 Uruguay Round of agriculture and services. Industrial tariffs will probably be included, but there was sharp disagreement between members on topics and objectives. Both the US and the Cairns Group of non-subsidising agricultural exporters sought large-scale cuts in agricultural support and protection, whereas the EU and Japan sought recognition of the 'multi-functionality' of agriculture (*e.g.* its contribution to the rural economy, the rural environment, food security, social rôle, *etc.*) and consequently the need for subsidy. On one hand, the USA seeks to remove obstacles to the sale of genetically modified crops and derived products, on the other hand, the EU has placed trade barriers on purported health and environmental grounds.

In an initiative to eliminate global tariffs on paper and wood products in MDCs by 2000 and LDCs by 2003, the WTO ran into opposition from the US Congress, on the grounds that it could encourage unsustainable and possibly illegal logging. The initiative stemmed from several sectoral trade liberalisation measures in the Accelerated Tariff Liberalisation package largely negotiated in the Asia-Pacific Economic Co-operation Forum, and will be discussed further in the November 1999 WTO meeting. As the world's principal trade forum, the WTO budget was tiny in relation to its ever-expanding responsibilities, and attempts were made to expand substantially the number of senior bureaucrats to pander to regional political sensitivities.

The fear of inflation receded towards the end of 1998 and the beginning of 1999. Price rises eased and global inflation decreased from 3.1% in 1997 to 2% in 1998. The decline in consumer and commodity prices then raised the spectre of deflation coupled to recession. Commodity prices as a whole fell by 25% during 1998 and were at their lowest for more than two decades. The price of non-fuel commodities was

at its lowest since 1986. Foodstuff prices fell by 9% in the year to October, and primary indications are that with good harvests the decline will continue. Short supply tended to shore up cocoa prices, and sugar prices stabilised. There was downward pressure on coffee prices.

Even though their economies were overshadowed by the world-wide economic and financial crisis, 11 of the EU countries (Austria, Belgium, Finland, France, Germany, Ireland, Italy, Luxembourg, The Netherlands, Portugal and Spain) replaced their national currencies with a new currency, the euro, at the beginning of 1999. Foreshadowed by R Mundell, the eminent economist who is still influential in international macroeconomics, the euro superseded national currencies and the straitjacket of relative rates when the euro was constituted. Full circulation of the currency is scheduled to take place in 2002. The launch of the euro was thought by some to be the final stage of economic and monetary union (EMU), and the IMF designated these countries as the 'euro area'; other terms widely used with optional hyphenation included 'eurozone' and 'euroland'. Monetary control moved to the 17 governing members of the European Central Bank (ECB) which now sets a common, single interest rate for all 11 countries. The internal exchange rate of the euro to the 11 national currencies was fixed irrevocably. A six-member executive board chaired by W Duisenberg shared decision-making with the central bank governors of the 11 member countries. The stated status of the ECB was one of independence and neutrality, with price stability and low inflation as priorities. Some bankers and economists were perplexed by (i) the fact that the ECB employed only 500 staff while the national central banks employed over 60,000, (ii) the governors of the national central banks held a majority of votes in the governing council (11 votes against the ECB's six), (iii) historical mishaps of highly decentralised central banking systems *e.g.* the original Federal Reserve System of the USA, (iv) the lack of rôle as lender of last resort, (v) member states operating differing political and economic policies, and (vi) the impact of e-commerce. Thus, the ECB could be in danger of becoming an emasculated secretariat. Even the European System of Central Banks, the decentralised network comprising the ECB and national banks, would not have an explicit rôle as lender of last resort.

Export-led growth in output in the euro area during the first quarter of 1998, followed by reasonable levels of domestic demand, helped launch the euro with an air of optimism. During the first half of 1999, however, the declining value of the euro against the dollar and sterling created political as well as economic difficulties for the proponents of incorporating the UK into the euro currency zone. Unemployment, at more than 15% of the labour force in the EU as a whole, remained a politically sensitive issue for the dominant left-of-centre parties that controlled the European Parliament and most of the member countries. Nevertheless, in the first half of 1999, there was a surge in issues of euro-denominated debt and all top 10 issues exceeded $1 billion. This would indicate a deepening of the liquidity and breadth of euro-dominated fixed-income debt. Denmark, Greece, Sweden and the UK did not adopt the euro, but retained the flexibility to join at a later date. The European Commission (EC), consisting of 20 Commissioners and which initiates and implements EC legislation and is guardian of EC treaties, came under close and critical scrutiny especially in relation to the EC Humanitarian Office. In April 1999, the Commission resigned en bloc, in the wake of allegations of financial mismanagement, secrecy, failure to adapt from a policy culture to a management culture, and an unwillingness to accept responsibility. The Commission employed around 15,000 civil servants in 1998. To widespread disapproval, members of the European Parliament rejected a move to link their expense claims to the money actually spent on travel.

In October 1997, the UK Government set out five tests for UK membership of the eurozone. These tests have been criticised as being too subjective. Bodies such as the Institute of Directors (IoD) proposed quantifiable economic tests, arguing that the eurozone should adopt an Anglo-American economic model for convergence between the UK and eurozone economies. The IoD tests included (i) a requirement for eurozone unemployment levels to fall to UK levels; (ii) the gap between UK and eurozone taxation shares should halve but without any increase in the UK's overall level of taxation; (iii) the proportion of fixed-rate mortgages by UK homeowners should converge with that in the eurozone; the fact that about 75% of UK mortgage debt is held at variable rates means that the UK economy is acutely sensitive to changes in base rates (according to the Centre for Economics and Business Research, London is four times as sensitive to interest rate changes as the rest of the UK), and thus a 'one size fits all' monetary policy operating in most of Europe would damage the UK; (iv) the eurozone should account for more than 50% of UK current

account earnings rather than the 43% level in 1998-1999; (v) the GDP correlation coefficient between the UK and euroland should exceed that between the UK and the USA for a decade; at present, the UK and USA business cycles are closely aligned but there is little relationship between growth in the UK and that in the eurozone. No constitutional tests were devised.

A major strategic document, Agenda 2000, included measures to reform the EU budget and its major spending policies, particularly the Common Agricultural Policy (CAP) and help for the economies of the less-developed member states. There was a movement away from production-related support, concomitant with the introduction of an integrated approach to rural development and agri-environmental measures. In the arable sector, a 20% cut in the cereals intervention price was proposed, alongside a single, non-crop-specific area-aid payment of 66 ecu *per* tonne for all eligible crops and set-aside areas. The Agenda 2000 package included a proposal for a new regulation on the financing of the CAP from the European Agricultural and Guarantee Fund, replacing Council Regulation (EEC) No 729/70. Denmark, Italy, Sweden and the UK (the 'London Club') pressed for abolition of the 14-year-old EU milk quota system which restrains output and competitiveness. The highly regulated milk quota system was set up in 1984 in response to growing 'mountains' of surplus butter and skimmed milk powder. Germany and France were hard-line opponents to reform of the quota system. The Agenda 2000 reforms were designed to allow the EU to accommodate enlargement with new member states from Central and Eastern Europe, and from the Mediterranean region. In a Heads of Government meeting in June 1998, the EU agreed that negotiations should begin with Cyprus, the Czech Republic, Estonia, Hungary, Poland and Slovenia. Countries such as Bulgaria, Latvia, Lithuania, Romania and Slovakia were required to carry out economic and political reforms before accession negotiations could begin, and were therefore invited into partnerships with the EU. Membership of a powerful trading bloc was attractive to isolated countries or groups of countries with weak economies, especially during the formative years of the WTO.

Institutional and constitutional reforms to the EU were contentious issues. These included subsidiarity of decision-making by majority vote in the EU Council of Ministers; reweighting of votes according to population size; possible strengthening of the EU Common Foreign and Security Policy, including refreshing the linkage between NATO and the Western European Union, the security and defence organisation of the EU; setting up a transatlantic free-trade area with the USA; and so-called unfair tax competition.

Existing and potential commitments of the public sector to social security or protection budgets, and the efficiencies of the institutions that deliver and monitor benefits, were of concern to all nations. Reviews of measures to stimulate employment, reduce benefits, upgrade taxation systems, modify child welfare schemes, and improve health were commonplace.

Created at Bretton Woods, USA, in 1945 by 44 countries, the IMF was designed to foster monetary co-operation by enforcing strict rules of behaviour in a world based on the gold standard and fixed currency-exchange rates. Following the abandoning of the gold standards by the USA in 1971, the system of fixed exchange rates collapsed, forcing the IMF to concentrate on providing advice and information to its members, which in 1998 numbered 182 countries. Lately, the IMF has assumed the rôle of international lender of last resort to support long-term efforts at economic reform, providing credit lines, other facilities and imposing of austerity plans. It is not a bank *per se*, but provides temporary financial assistance by selling a member's special drawing rights or other members' currencies in exchange for the member's own currency. The member can then use the purchased currency to alleviate its balance of payments difficulties. Nations in Latin America, Africa, Asia, and Central Europe have been assisted in restructuring their economies since the 1982 debt crisis. Criticisms of the IMF included its secretive dealings, lack of democracy in its composition, poor responsiveness to the needs of poorer countries, and the tendency to bail out international investors rather than the economy of the recipient country. In reality, even with a fortified capital base, IMF assistance can only be modest in an era when financial flows are dominated by thousands of banks; securities firms; and mutual, pension and hedge funds all able to move capital across borders electronically. To this must be added a vast array of financial instruments available in the international arena, making the co-ordinating rôle of the IMF very difficult. At the IMF's key ministerial committee meeting in October 1998, the World Bank and the IMF presented an updated version of the 1989 'concordat' drawn up after the public row following the loan of $1.25 billion by the Bank to Argentina,

against the advice of the IMF. Many regarded the response of the Bank to be too slow in promoting financial Sector reform, and the IMF too hasty. The Group of Seven leading industrial countries insisted that the Bank should develop an 'emergency capacity' for focusing on financial sector reform and support for vulnerable groups in society.

According to a report in April 1999 from the World Bank, net flows of overseas aid to LDCs have fallen to their lowest level in real terms since 1981, with little sign of a significant recovery in prospect. The total debt of LDCs reached $2,500 billion, but total grants fell to $23 billion.

International travel and tourism has become a huge industry. Earnings were expected to exceed $450 billion in 1998 and involve at least 620 million arrivals involving about 300 million individuals. For many economies, tourism is now a prime industry, and agricultural holdings have attempted to diversify into hosting tourists and visitors to offer exposure to a rural existence, exploiting the visual amenity and local history. Nearly 70% of users of the World Wide Web were thought to have accessed a travel-related site in 1998 (a large proportion, too, accessed health-related sites). Realisation of the rôles and responsibilities of heritage sites, the rural countryside, tourist facilities and transport were beginning to exercise governments. For those sensitive to the environmental impacts of travel and tourism, there was growing awareness of the need to preserve the natural world and human cultures – leading to so-called 'ecotourism' which should be sustainable compared with the bulk of present-day travel and tourism. In theory, ecotourism could and should have benefits for travel, conservation, habitat, maintenance and employment, as well as raising the general level of environmental and cultural awareness. The establishment of bodies such as The Ecotourism Society might aid in formulating action plans and programmes to foster sustainable travel and tourism.

Following the high growth reported in 1997, when demand rose by 6%, growth of the textile market in 1998 was barely detectable in economic surveys, accounting for an excess of capacity in the textile industry and a consequent dramatic fall in prices and profitability. The Asian economic crisis caused market volatility but improved the competitiveness of those exporting nations at the nub of the crisis. In 1998, 48.6 million metric tonnes (mmt) of textiles were produced, including 19.2 mmt of cotton-based textiles.

One of the most dramatic price falls of recent times for energy caused financial turmoil for the oil industry in 1998-1999, putting pressure on oil exporters and inducing mergers of some of the largest corporations and companies in the industry. Not all the members of the Organization of Petroleum Exporting Countries (OPEC) initially abided by agreements made during 1998 to take part in a world-wide round of production cuts, irrespective of the extent of the global supply surplus and the fall in demand from Asia. By the spring of 1999, however, OPEC members reduced production, and oil prices started to rise.

Global demand for natural gas, the least polluting fossil fuel, continued to grow faster than that for oil in 1998. The International Energy Agency estimated that demand for gas rose by 2.6% per year, compared with 1.9% for crude oil. Research, development and marketing devoted to low-cost methods for converting natural gas into low-pollution diesel and other middle-distillate fuels, including kerosine (paraffin-oil), were somewhat impeded by low oil prices.

By 1998, world coal consumption had grown to the equivalent of 20 million barrels of oil a day. This increasing reliance on coal reflected a move away from nuclear power and the greater use of low-cost coal. For the twelfth consecutive year, world-wide coal consumption exceeded five billion short tons; both the USA and China produced more than one billion short tons of coal annually. This cost-efficient preference for coal undermined international efforts to reinforce the Kyoto Protocol on Climate Change and the Global Environment Facility, and was the product of not applying environmental accountancy factors.

At the beginning of 1998, there were 437 operational nuclear power units in 33 countries, compared with 442 a year earlier. On the other hand, the total operating capacity was 351,795 MW, an increase of 831 MW over 1997, according to the International Atomic Energy Agency. World-wide, nuclear power units produced a total of 2,276.32 TWh. Thirty six units were under construction in 14 countries. The total number of commercial power reactors permanently shut down reached 80; decommissioning and disposal costs have yet to be calculated.

There was tangible evidence that low prices for fossil fuels started to weaken investments into alternative energy sources, but according to the Worldwatch Institute in Washington, USA, the capacity for generating wind power reached 7,630 MW, compared with

10MW in 1980. Shipments of photovoltaic solar cells rose 43% in 1997 to 126 MW. According to the International Energy Agency, renewable energy sources, mainly in the form of hydroelectricity and biomass (*e.g.* firewood, crop waste, charcoal, animal waste *etc.*) supplied between 15-20% of the world's energy demand. The world biomass production was calculated to be 6.9×10^{17} kcal *per* year, of which only 7% was utilised. The fact that in many countries conventional fuels were subsidised directly or indirectly, that there were insufficient incentives to convert to alternative energy sources, and that there were only small investments in research and development (R&D) in this area by both the public and private sectors, meant that technological progress and the numbers of commercially successful schemes were extremely disappointing.

The Annual Corruption Perception Index, produced by Transparency International on 85 nations, ranked Colombia, Indonesia, and Nigeria as the world's most corrupt large countries. Canada, Denmark, Finland, New Zealand and Sweden were perceived among the least corrupt.

Populations

The world population was estimated by the Population Reference Bureau to be 5.926 billion by mid-1998, representing an increase of 86 million over the previous year and indicating that by mid-1999 the global population would probably exceed 6 billion (the 'Y6B' phenomenon). The annual rate of increase of approximately 1.41% was less than the 1.47% estimated for the previous year reflecting the decline in birthrate of many LDCs. At the 1999 rate of increase, the world's population would double in 49 years. As a result of deaths due to AIDS, however, and to changing social trends, the UN Population Division forecast a world population of 8.9 billion by 2050, eventually stabilising at just under 11 billion in 2200. Of the 137 million children born in 1998, 2 million fewer than in 1997, more than 90% were born in the LDCs. About 53 million people died in 1998; 78% of the deaths took place in the LDCs. Demographic studies showed that 32% of the population was below the age of 15 in 1998, but the figure was 37% in LDCs outside China and 44% in Africa. For MDCs, the population below age 15 fell to 19% from 20% the year before. This situation of an ageing population resulted from the low birthrates in Europe and Japan, raising concerns over future welfare, taxation and societal pressures.

The percentage of the world's population living in urban areas rose to 44% in 1998 from 43% in 1997. In LDCs, however, 36% of the population was urban-based contrasting with 73% in MDCs. The usual definition of urban population includes those living in towns of 2,500 or more inhabitants, cities, and provincial and national capitals.

Life expectancy at birth in 1998 was 64 years for males and 68 for females; in MDCs the figures were 71 and 79, and for LDCs, 62 and 65. Infant mortality rates varied from five infant deaths per 1,000 live births in western Europe (Finland had the lowest rate of 3.5), to 64 in some LDCs.

The total population of LDCs in 1998 stood at 4.666 billion, 82 million more than in 1997, representing a 1.73% increase per year, 1.99% for LDCs outside China. Women averaged 3.9 children each in LDCs outside China, far from the 'two-child family' needed to stabilise the size of the world population. The annual growth rate of population in Africa was estimated to be 2.5% in 1998, the world's highest, and if sustained would lead to a doubling of the continent's population in 27 years, from its current level of 763 million. Life expectancy at birth was 50 years for males and 53 for females; infant mortality was the world's highest at 91 infant deaths *per* 1,000 live births. In some areas of Africa affected by HIV/AIDS, life expectancy was less than 40 years. At 500 million in 1998, the population of Latin America had increased by 1.8% from the previous year. Women averaged three children, ranging from 1.4 in Cuba to 5.1 in Guatemala. The infant mortality rate was 36 in 1998, down from 39 in 1997. Life expectancy was 66 years for males and 72 for females. The population of Asia was estimated at 3.604 billion, an increase of 54 million over 1997. The population growth rate declined to 1.5% in 1998 from 1.6% in 1997, attributable to a small drop in the growth rate in India. Women averaged 2.8 children in 1998, but 3.3 in countries excluding China in which women averaged 1.8 children. In India, women averaged 3.4 children. Life expectancy in 1998 was 64 years for males and 67 for females.

In 1999, the UN Population Division reckoned that the population of India reached one billion, and was projected to overtake in four decades the population of China, currently at 1.248 billion. Its urban population has quadrupled in half a century. According to the Worldwatch Institute, half of India's adults are illiterate, more than half its children are undernour-

ished, and a third of its people live below the poverty line. Food production has barely kept up with population growth, and underground water reserves are being used twice as fast as they are being replaced.

FAO estimated that over 800 million people lack adequate food. Protein and micronutrient deficiences were expected to become increasingly serious, particularly for women and children. More than 33% of those living in sub-Saharan Africa were predicted to be food-insecure by 2010. More than 75% of the world's poor live in rural areas in LDCs, reflecting an historical legacy based on political discrimination, inadequate resources to derive livelihoods, pests and diseases of crops and livestock, social instability, ill-health, and utilisation of natural resources (soil, water, forests, fisheries, grazing areas) beyond their replenishment capacity. As was pointed out in the 1998 Third System Review of the Consultative Group on International Agricultural Research (CGIAR) entitled *The International Research Partnership for Food Security and Sustainable Agriculture*, addressing the nutritional needs of the human population remains one of the most important challenges of the future. Escape from the Malthusian trap is temporary. The international community cannot and must not accept the persistence of extreme poverty and deprivation in the midst of unprecedented prosperity. World agriculture was seen to be linked to five environmental threats: (i) water scarcity, (ii) soil degradation caused by such factors as salinisation, nutrient depletion, erosion, *etc*., (iii) loss of global biological diversity, (iv) the effects of global climate change and greenhouse gases, and (v) persistent trends of desertification. All of these threats are compounded by the expansion of cultivated land and by farming intensification using conventional methods. The CGIAR saw its main rôle as participating in technological developments such as genomics; bioinformatics; genetic transformation of crops, trees, livestock and fish; molecular breeding; molecular diagnostics of pathogens; vaccines; the Internet; computing to process large-capacity databases; remote sensing; new management systems such as artificial intelligence models, strategic forecasting, negotiation models, *etc*. It was pointed out that the total bilateral and multilateral assistance to agriculture in LDCs amounted to $10.3 billion in 1995, 20% below aid levels in 1991. Estimated agricultural subsidies were in the order of $335 billion in MDCs.

In the MDCs, the population in 1998 was 1.178 billion with a growth rate of just 0.1%. Without population expansion in the USA (from 267,636,000 in 1997 to 270,029,000 in 1998), MDCs would have recorded zero or even a negative growth rate. Thus, in Europe in 1998 there were more deaths than births, and 13 European countries experienced population declines. The Czech Republic, Italy, Latvia, and Russia had the world's lowest fertility of 1.2 children each. Life expectancy at birth in Europe, including the European republics of the former Soviet Union, was 69 for males and 77 for females. In 1998, about 17% of the population of Sweden was aged 65 or older. Japan's population was ageing faster than any other country and was projected to reach a level of 24% aged 65 or older by 2025. Increasing numbers of people aged 85 or older will have serious implications for taxation, welfare benefits, healthcare, and social structures, as well as for the design, planning, and management of housing and communal environments. In much of Europe and in the USA, about 5% of the population aged 65 and older resided in nursing homes.

The total number of people of concern to the Office of the United Nations High Commissioner for Refugees (UNHCR) stood at 22.3 million at the beginning of 1998, *i.e.* one out of 264 living persons in the world. These displaced people included 12 million refugees, 3.5 million returning refugees in the early stages of reintegration, 900,000 asylum seekers, and 5.9 million internally displaced people, *i.e.* in a refugee-like situation but who had not crossed an international border. Although repatriation was the preferred solution for most victims of conflict and for those countries acting as temporary hosts, most refugees in 1997 and 1998 returned to countries still embroiled to various extents in warfare.

As in the previous few years, civil unrest in the Great Lakes region of Africa and the surrounding countries provoked large-scale population movement. Refugees fleeing the conflict in the Democratic Republic of the Congo (formerly Zaire) moved to Angola, Burundi, the adjacent Republic of the Congo, and Tanzania where 260,000 Burundians comprised the regions largest single refugee group. In the West African region, there were prospects for the establishment of the rule of law in 1998. Around 50,000 refugees returned from Côte d'Ivoire and Guinea to Liberia, and 135,000 refugees were repatriated to Mali and Niger. In Sierra Leone, however, 200,000 refugees crossed into Guinea and 55,000 into Liberia, such that by 1998 over 450,000 Sierra Leoneans were living as refugees in neighbouring countries. Problems still afflicted East Africa and the Horn of Africa. Dis-

agreements were flaring up between Ethiopia and Eritrea. Civil war continued in The Sudan. Even so, 70,000 Ethiopians were repatriated from The Sudan, and 30,000 refugees returned to Somalia. In Southern Africa, a new wave of refugees left Angola.

In Asia, a combination of fighting and violations of human rights, especially those of females, prolonged the misery experienced by the population of Afghanistan. Despite the troubles, over 80,000 Afghan refugees returned from Pakistan and over 2,000 from Iran. Conflict in Cambodia in 1997 and 1998 drove out 70,000 refugees to Thailand. More than 800,000 persons were internally displaced from the Jaffna peninsula in Sri Lanka and became dependent on humanitarian assistance in 1998. About 10,000 refugees from Myanmar (Burma) remained in Bangladesh following their flight in 1991-1992, and about 100,000 ethnic Karen and Karenou refugees were displaced to the border with Thailand. With UNHCR assistance, Nepal accommodated 93,000 Bhutanese refugees.

In Europe, more than 1.8 million people remained displaced in and outside the former Yugoslavia. This was counterbalanced by 120,000 people who were repatriated to Bosnia and Herzegovina in 1997 to relatively safe areas *i.e.* where their particular ethnic group was in the majority or ascendancy. The crisis in Kosovo had displaced nearly 300,000 Albanian refugees by the end of the year, with mass movement starting to take place into Albania, Macedonia and Montenegro, creating conditions that would give rise to another humanitarian crisis. Pressure started to develop on EU countries to absorb the refugees. In Russia, it was estimated that at the beginning of 1998 there were 4 million forced migrants, 1.2 million of whom were registered with the Russian Federal Migration Services. Displacements in recent times came from fighting in the Chechen Republic and Abkhazia in Georgia.

Agriculture and Food

The predominant factors influencing world agricultural markets in 1998-1999 were the economic disturbances occurring mainly in Asia, a continuing downward pressure on commodity prices, the aftermath of the El Niño event on agricultural productivity, drought-induced and warfare-related food emergencies, decreasing food aid, new technologies, the growing influence of the World Trade Organization, and the declining political and economic influence of agriculture in developed countries. For growers in general, the decline in farm income was relatively severe, and had serious effects on those businesses that were dependent on inputs to agricultural production. Industries downstream of agriculture benefited from low prices.

Analyses of agricultural and food production in 1998 by FAO; (see http://apps.fao.org) reveal that total (world) agricultural production declined by 0.2% from the 1997 record-high level, with the decline most marked in developed countries, whereas a rise took place in less-developed countries (LDCs). World food production remained constant, but, again, there was a rise in food production in the LDCs such that it more or less kept pace with the rise in their population levels, accounting therefore for a barely detectable decline in *per capita* food production. In developed countries, however, there was a marked decline of about 2% in total food production and a similar decline in *per capita* food production. Food emergencies and shortages in 1998 occurred in Central and East Africa, Indonesia, Iraq, North Korea, Russia, The Sudan, and the former Yugoslavia. According to FAO, food aid in the form of shipments of cereals had dropped from 9.44 mmt in 1994-1995 to 5.30 mmt in 1996-1997, declining further to an estimate of 5.34 mmt in 1997-1998. Both the USA and the European Union (EU) had reduced their cereal food aid since 1992-1995 by 70% and 77% respectively, effectively shrinking governmentally controlled grain stocks and reinforcing policy shifts to reducing production price supports.

Fortunately, the Asian economic crisis that by the beginning of 1998 had afflicted Thailand, Indonesia, Malaysia, South Korea, the Phillipines, Singapore, and to a lesser extent, Hong Kong and Taiwan, did not have as large an effect on agriculture as originally predicted. Presumably this was because none of the countries was a large agricultural trading nation. In any case, sales of agricultural commodities are usually negotiated well in advance of shipments, and nations such as Indonesia, South Korea and Thailand were recipients of credits from exporting nations and international agencies.

As the impact of economic problems spread during 1998 to Brazil, Chile, China, Japan, Mexico, Russia, and Venezuela - most of which represent large import markets as well as being significant exporters of some agricultural goods - it was inevitable that there was a sharp decline in agricultural prices that continued well into 1999. The growing realisation of the ability of

agricultural and food technology to adapt rapidly and efficiently to market-driven demands, and to address shortages, also suppressed market prices, favouring large-scale, efficient and competitive operations, especially those with ownership of marketable intellectual property such as protected cultivars favoured by customers of the primary produce. An inability to capitalise on value-added processes and little market 'muscle' at the consumer or customer interface meant that the profitability of agriculture remained in the doldrums, except for those concerned with niche markets, or operating in subsidy-dependent regime, or vertically integrated with downstream companies. While commodities such as cocoa, coffee, cereals, oilseeds, sugar and rubber traded at around historic low levels, future contracts attracted increasing interest, lifting volumes to record levels as commodity users took the opportunity to lock in low prices for the future. Commodities are regarded as a major vehicle by investors to hedge against the effects of inflation and other possible long-term developments. Depressed commodity prices aid governments of industrialised countries in suppressing inflation.

FAO estimated that the value of agricultural exports from LDCs fell by $4.6 billion in 1998, with sugar, cotton and rubber accounting for 85% of that decline. Earnings from agricultural raw materials fell by 23%, equivalent to $2.5 billion in 1998, with cotton and rubber, the largest commodities in the raw materials group, accounting for most of the decline. In contrast, oilseeds and beverages fared better. The value of oilseed exports fell by only 1%, whilst gains in cocoa and tea offset falls in coffee prices.

The crop protection industry in 1998-1999 underwent further changes with mergers, acquisitions and strategic alliances. Many companies continued the trend towards greater specialisation and outsourcing of functions, especially in R&D and manufacturing of active ingredients, paralleling the trends in the pharmaceutical industry. Herbicides maintained a dominant rôle in crop protection in MDCs. Around 80% of the crop protection market was accounted for by the ten leading companies (in order of 1997 sales: Novartis, Monsanto, Zeneca, DuPont, AgrEvo, Bayer, Rhône-Poulenc, Dow AgroSciences, Cyanamid and BASF). New combinatorial chemistry techniques, coupled to automation of biological screens and functional genomics, have revolutionised the search for new agrochemicals and other products by the agrochemical industry. Contrary to the impressions given by organisations campaigning against multinational companies, around 70% of the £23 billion global seed trade during the year was controlled by public-sector bodies; of the remaining 30% of the market there were about ten major international companies and numerous small-scale companies operating within countries. Nonetheless, the competition authorities are alert to possible market abuse, especially with the recent involvement of biotechnology and patenting.

In a pivotal paper in *Food Policy* (*23*, pp 371-381, 1998), David Wood critically analysed international agricultural research policy and the increasing emphasis on the rôle of 'ecological principles' in the development of farming systems. Agroecology and agroecosystems are commonplace terms, but the underlying widely accepted ecological principles do not fully take into account the following six factors. (i) Succession and the need for controlled ecosystem disturbance to maintain highly productive early successions. (ii) The fact that diverse agroecosystems are not invariably more sustainable and productive than less-diverse systems, and are demonstrably not more 'stable' than uniform stands (witness the pure natural stands of wild grass species and of conifers), compared with the fragile but highly species-diverse tropical rain forests and coral reefs. (iii) The definition of 'sustainability' which might relate to 'stability', 'resistance' or 'resilience' to forces which bring about change. Sustainability could infer a lack of adaptability or response to imposed stresses and desperately needed inputs to improve productivity. Rarely does the term 'sustainability' incorporate adequate economic survival and adaptability. (iv) Crop introductions, *i.e.* alien species, are crucial for developing countries; of 72 LDCs for which information was available, 50 (69%) relied on more than half of their domestic crop production on crops previously introduced from other continents. Enhanced performance usually follows introductions to a new environment, especially in the absence of pests and diseases, outperforming local crops and with minimal environmental risk when the introductions remain under farmer control. Oftenplace, new commodities can attract premium prices. (v) The poor must be able to benefit from and be involved in any new agricultural policy that affects low-cost food production. (vi) Any new agricultural policy must not adversely affect agriculturally marginal or wild habitats, by increasing the land area devoted to cropping. Under the guise of environmental protection, anti-technology groups appear willing to condemn the agrarian poor to the daily grind and vulnerability of subsistence agriculture. Thriving rural

economies need to be based on activities other than low-level primary production.

Cereals

The production of wheat, coarse grains (barley, maize, millet, oats, rye and sorghum) and milled rice was estimated to be 1886 mmt in 1997-1998 and was forecast to decline to 1,850 mmt in 1998-1999, according to the United States Department of Agriculture (USDA). Provisional calculations indicate that production was taking place on a rapidly declining area of cultivated land, indicating greater efficiency of production, such that more than 12 million hectares of land were taken out of cereal production during the past 3 years. In contrast to declining cereal production, the utilisation of cereals for both livestock feed use and for food and other use increased to 1,867 mmt in 1998-1999, with most of the increase attributable to wheat consumption. Ending cereal grain stocks were therefore forecast to decline from *c.* 322 mmt in 1997-1998 to *c.* 305 mmt in 1998-1999; again, the decline was most pronounced in wheat stocks, down to 124 mmt from 135 mmt in 1997-1998, but milled rice stocks fell to only 43 mmt, down from 52 mmt. As a percentage of utilisation, total cereal stocks declined slightly from 17% to 16% in 1998-1999, and represented 60 days supply or 16% of world annual consumption.

Oilseeds

In 1998-1999, the USDA forecast that the world production of the seven main oilseeds (soybeans, cottonseed, groundnut or peanut, sunflower seed, rapeseed, copra and palm kernel) would increase from 287.1 mmt in 1997-1998 to 290.8 mmt in 1998-1999, continuing the trend of increases which are forecast by *Oil World 2020* to continue over the next 20 years. Soybean (154.1 mmt), cottonseed (32.5 mmt) and copra (5.4 mmt) production declined from 1997-1998 levels but groundnut (27.6 mmt), sunflower seed (26.2 mmt), rapeseed (36.8 mmt) and palm kernel (5.4 mmt) levels increased. Oilseed ending stocks reached 25.4 mmt in 1998-1999 compared with 22.2 mmt the previous year. Palm oil, as opposed to palm kernel oil, has the potential to expand substantially its share of world production because of its much higher yield per hectare than any seed oil, and relatively low production cost efficiency. The debate in the EU on transgenic crops was starting to influence trading and sourcing arrangements, especially in relation to the high-protein meals derived from certain oilseed and related seeds.

Sugar

According to the USDA, global production of centrifugal (freed from liquid) sugar increased from 125.4 mmt in 1997-1998 to a forecast record level of 126.5 mmt in 1998-1999. Most of the increase was accounted for by greater production in Brazil, the Caribbean, India, South Africa, and Turkey. The main producing countries were the EU (18.0 mmt), India (16.8), Brazil (16.6), USA (7.3), Australia (5.4), Mexico (5.1), Thailand (4.2), Cuba (3.2), Poland (2.2), Guatamala (1.8) and Russia (1.2). World consumption was forecast to increase to a record 127.5 mmt in 1998-1999, leading to a decline of beginning stocks to 25.2 mmt, representing 19.8% of consumption.

Coffee

World green coffee production in 1998-1999 was forecast by the USDA to increase to a record 107.5million 60-kg bags, compared with 94.3million 60-kg bags in 1997-1998. Most of the increase was attributable to an increase in production by Brazil (23.5million 60-kg bags in 1997-1998 to 35.8million 60-kg bags in 1998-1999). There were fears that Brazil would exceed its quota set by the Association of Coffee Producing Countries as part of a plan to limit exports of coffee and support sagging prices. Brazil enjoyed a bumper harvest in the 1998-1999 season, with its exports helped by the devaluations of its currency, the Real. Production increases were also noted by Colombia, Ethiopia, Mexico, Peru, Uganda and Vietnam. Coffee was the mainstay of the HIV/AIDS-affected Ugandan economy, earning nearly 70% of export revenues and employing 80% of the rural workforce. According to the International Coffee Organisation, Uganda exported about 3 million 60-kg bags, 90% of it from low-yielding types of robusta bushes. World coffee trade in 1998-1999 as measured by exports, increased to 81.1million 60-kg bags from 71.5million 60-kg bags the year previously, and beginning stocks declined to 23.3million 60-kg bags from 28.8million 60-kg bags. Hurricane Mitch which swept through Central America, killing about 12,000 people, severely affected agricultural exports, especially those of coffee, which in recent years account for 10-12% of world coffee production.

Cocoa

Pivotal to the high-value chocolate market, world cocoa bean production in 1998-1999 was forecast by the USDA to be closely similar to that of 1997-1998 at 2.69 mmt, below the level of 2.72 mmt recorded in 1996-1997, and leading to a supply deficit for the sec-

ond year in a row. An increase in production was noted by the main producing nation, Côte d'Ivoire, to a level of 1.15 mmt, as well as by Brazil (0.17 mmt), and Indonesia (0.31 mmt). Reductions in production were recorded by Ghana (0.36 mmt), Nigeria (0.13 mmt) and Malaysia (0.10 mmt). Despite a production shortfall globally, inventories seemed to be comfortable and futures prices were weak.

Tea

According to the Economist Intelligence Unit in its 1999 report *World Commodity Forecasts : Foods, Feedstuffs and Beverages*, high stocks and increasing supplies depressed prices of tea (*Thea sinensis*). A large surplus carried over from 1998 resulted from the fact that in 1998 black tea supplies exceeded 2m tonnes for the first time, with record production in Bangladesh, India and Kenya, and an upturn in Indonesia. Output in 1999 was expected to be lower as a result of dry weather. World tea consumption was expected to increase by 2.7% in 2000, to more than 2m tonnes, and by a further 3.5% in 2001. Nevertheless, tea imports in 1999 were running less than the 1998 level as European buyers used up existing stocks and Russian imports were retarded by its economic woes. All the main tea producers have the capacity annually to meet demands.

Mate, the 'green tea' of South America, produced by an infusion of the leaves and green shoots of *Ilex paraguayensis*, faced a production glut in Argentina, with production reaching 300,000 tonnes in 1998. Brazil, the world's second largest producer, harvested 170,000 tonnes in 1996. Exports have been targeted at the Middle East, Europe, the USA and Asia, with emphasis in advertising mate as a premium medicinal tea.

Cotton

With the area of land planted to cotton expected to fall to less than 33m hectares, output of cotton in 1998-1999 was forecast by the USDA to decline to 84.2 m 480-lb bales (approximately 18.6 mmt), from 91.4 m 480-lb bales in 1997-1998. Declines in production between 1997-1998 and 1998-1999 are expected to take place in the USA (18.8 falling to 13.5m 480-lb bales), China (21.1 to 18.8m 480-lb bales), and in the former Soviet Republics and Africa. Production in other major cotton-producing countries increased (Australia) or remained essentially unchanged (India, Pakistan and Uzbekistan). Consumption was expected to stagnate at 88.3m 480-lb bales (*c*. 19.2 mmt), influenced by demand for other natural fibres and synthetics as the fashion market changes. Increases in cotton consumption were noted in Brazil, Mexico, Pakistan and Turkey, but declines were recorded by China, Indonesia and the USA. Prices held firm in Australia and the USA, but the US government marketing subsidy of $700 million was due to run out in early 1999 and the withdrawal of funding was expected to hit the cotton futures markets.

Rubber

In view of the fact that almost 75% of the world's natural rubber production came from Southeast Asia, the Asian economic crisis had a serious impact on the rubber industry in 1998. It was not possible to stabilise rubber prices as outlined in the International Natural Rubber Agreement between producer and consumer countries. Although the International Natural Rubber Organization (INRO), designed to implement the agreement, was able to make strategic purchases late in 1998, Malaysia (the third longest rubber-producing country) threatened to withdraw from INRO, and Thailand (the second largest producer) indicated likewise. It was only through political instability that Indonesia, (the world's largest producer nation) did not follow suit. Both Malaysia and Thailand proposed that the Association of Natural Rubber Producing Countries would oversee a cut in production and establish a buffer-stock mechanism. There was a slowing in the growth rate of rubber consumption to 2% in 1998, whereas world synthetic rubber consumption was 3.8% higher. Legislation and litigation in the USA was stimulated by reports of allergies to powdered natural-rubber latex gloves used by the medical profession. By mid-1998 more than 125 cases were pending in various state courts.

Tobacco

According to the World Tobacco File, there was a further decline in cigarette consumption in 1998, which began in 1997 when global consumption of 5.2 billion cigarettes had fallen by 0.4% from the previous year. The multinational manufacturers faced the hostility of anti-smoking groups, declining profits, and the potential of huge legal costs and settlements associated with the effects of smoking on human health. In the USA, a federal appeals court ruled that the US Food and Drug Administration had no authority to regulate cigarettes as if they were drugs. Tobacco farmers suffered from lower prices as leaf purchases by the manufacturers were markedly reduced. In contrast, the use of molecular technology to modify tobacco to produce valuable medicinal and veterinary products attracted considerable attention.

Pepper

The International Pepper Community (IPC), a UN-affiliated organisation, estimated in 1998 that world pepper production for 1999 would reach about 200,000 tonnes, up 5.8% from 189,000 tonnes in 1998. IPC calculated that 138,799 tonnes of pepper were exported in 1998, and 80,444 tonnes in the first half of 1999. India, the largest black pepper producer, had over-optimistic estimates of its production downgraded to 70,000 tonnes in the 1998-1999 season; 60,000 tonnes were produced the year before. For Vietnam, pepper has become an important export crop, earning about $70 million in 1998 for about 14,000 tonnes exported. Brazil, China and India are also major producing and exporting nations. Demand for pepper, which is used in food preparation, medicines, cosmetics and perfume, is expected in the short to medium term to exceed supply in a normal production year

Wood, Paper and Pulp

Politically expressed environmental concerns about timber harvesting virtually stopped all new timber sales from national forests in the USA during 1998. Without addressing the biodiversity issue, some claimed that the reduction in harvest would render the forests more liable to catastrophic fires because dead or dying timber would generate a reservoir of inflammable material. Lumber production in the non-Federal forests, however, increased to meet the strong housing market. Eastern European production of lumber increased, but it was not thought to be adequate to meet demand. A decline in timber harvests and lumber production was recorded in Russia. Logging in the Yangtze River watershed in China was brought to a halt because of heavy summer floods. During the third quarter of 1998, lumber exports, notably hardwoods, began to improve as the major Asian economies showed signs of improvement.

Paper and paperboard production almost reached 300 mmt by the beginning of 1998, increasing by 5.8% over the 1996 level. At 28.9% of the total, the USA remained the largest producer nation. Although data were not available at the time of preparing this article, it would be unlikely that there would be any increase in production levels during 1998 in the light of the Asian economic crisis. In 1997, the largest increases in paper, paperboard and pulp production were reported by Indonesia. In 1998, however, Indonesia's economic woes, and the long dry season followed by large-scale forest fires, would have caused sharp decreases in production. In Europe, there were rises in paper, paperboard and pulp production in Finland and Sweden. Paper and paperboard production increases were declared by Belgium, France, Germany and Italy. Financial results world-wide in 1997, the latest date for figures, were well below the record levels set in 1995.

At the end of 1998, import controls were imposed in the UK on Chinese-produced timber and wood-based packaging in an attempt to inhibit further the invasion of the tree-killing beetle, *Anoplophora glabripennis*, which may have already become established in the UK. The UK Forestry Commission established an ambitious programme to make the UK more self-sufficient in timber and wood products, increasing annual volume from about 9 million m^3 to 15 million m^3 by 2020. This will require investments in plantation and processing facilities of the order of £2 billion. At present, about 85% of the timber and wood products used in the UK is imported, at a cost of around £8 billion.

Food Processing, Retailing and Consumer Issues

Increased demand was noted in 1998-1999 for convenience foods, vegetarian products, 'functional foods', and 'natural' or 'organic' produce, a trend noted over several years and strongly influenced by advertising and frequent favourable, if unscientific, reports in the media.

Food poisoning incidents maintained their high levels in most parts of the world, irrespective of government-led advice on food hygiene, new methods of detecting pathogenic organisms, greater involvement of commercial catering services, well-established as well as new methods of food sterilisation, and the availability of improving sanitation conditions for a large portion of the world's population. The huge costs of around £4 billion to control the bovine spongiform encephalopathy (BSE) outbreaks in the UK, have made the BSE crisis the UK's most expensive peacetime catastrophe, which will undoubtedly echo in the international beef and other livestock trade for decades, as well as influencing the utilisation of livestock feedstuffs such as cereals. Largely confused with food poisoning and contamination, mainly as a result of simplistic and alarmist publicity by pressure groups, the issue of genetically modified (or manipulated) crops and food led to consumer resistance in Europe, and in the UK in particular. See also Plant Biotechnology p. 38.

The Asian economic crisis had a deleterious effect on food exporters to those countries most affected, but globally the food industry showed remarkable resilience, most notably in Latin America.

Marketing identities through advertising and packaging, including the introduction of plastic bottles, dominated the beer scene in 1998-1999. Company consolidations and takeovers continued. Distilling companies continued to try to enter new markets such as the 'cocktail culture' for consumers aged 18-25, and flavoured spirits. Most oenophiles considered that the 1998 vintage was generally good for all wine-growing areas and with the exception of auction markets, which were affected by the Asian economic crisis, prices continued to rise. New consumers ensured that strong demand for wines was maintained. Southern Hemisphere wines enjoyed expanding markets. Highly competitive markets for soft drinks, particularly for the two dominant forces of Coca-Cola Co. and PepsiCo Inc., led to the development of a litigious edge in their interactions. Many soft drink companies sought to diversify their product range. Regulators in the USA approved two new synthetic sweeteners, sucralose and acesulfame-K, which could assist in revitalising the low-calorie 'diet' segment of the market. G Inglett of the USDA developed the fat substitute 'Nu-trim' released during the year. Prepared by thermal and mechanical shearing of oat or barley flour, leading to the formation of a hydrocolloid rich in beta-glucans, the product was launched as a fat replacer in baked goods, ice-cream, non-dairy cheeses, *etc*. Also launched was the Proctor & Gamble Co. product 'olestra', a no-calorie, no-cholesterol fat substitute.

Ethical and health issues were foremost in the concerns expressed by consumer organisations about the introduction of genetically modified (GM) foods. In May, a committee of the Codex Alimentarius Commission (CAC, the Rome-based UN body responsible for setting international food standards) held a meeting to discuss food labelling. Consumer groups and other NGOs urged the committee to require labelling of all GM food. Consumer International, a federation of 235 consumer organisations in more than 100 countries, ran a campaign to contact the committee and demand mandatory labelling. The committee sensibly decided, however, to seek further expert scientific opinion and review the issue later in 1999. CAC approved in 1999 the establishment of an inter-governmental task force charged with accelerating the development of standards for GM foods. CAC hoped that the standards will be prepared and adopted by 2003.

The 'organic' food standard developed by FAO/WHO Food Standards Programme took into account regulations operating in several countries and the standards applied by producer organisations. Organic agriculture (see p. 60) is assumed to emphasise the use of management practices in preference to 'off-farm inputs', and uses 'cultural, biological and mechanical methods, as opposed to using synthetic materials'. Even though it has enormous potential for organic and low-input agriculture and horticulture, GM technology illogically is not accepted under current CAC guidelines. The draft EU regulation amending Council Regulation 2092/91 on organic standards proposed a complete ban on the use of GM technology.

Organic farming, on the one hand, raises risks of faecal contamination not only of foodstuffs but also of waterways, food poisoning, high levels of natural toxins (*e.g.* aflatoxins) and allergens, contamination by copper- and sulfur (contaminated with lead)-containing fungicides, production of blemished, diseased and irregular produce of low consumer and food processing acceptability, low productivity, and creation of reservoirs of pests and diseases, including sources of weed propagules. On the other hand, regenerative agricultural management systems based on organic fertiliser can preserve carbon and nitrogen in the soil, thereby reducing greenhouse-gas emissions, dispense with synthetic agrochemicals, and sustain on-farm, in-field biodiversity. In Europe, where the middle classes spend less than 15% of their income on food, and may even expect organic agriculture to be subsidised, and suffer from a surfeit of foodstuffs, organic agricultural systems have viable market niches. In LDCs, however, where more than 70% of income is spent on food, and production perturbations caused by pests, diseases, weeds and bad weather lead to starvation and even suicides, there is a profound requirement for improving agricultural productivity and efficiency, and biotechnological approaches, including GM crops, are real options. The marketplace will ultimately determine the success and scale of organic farming enterprises, perhaps rapidly after the furore on GM crops has subsided. Competition between organic farming units will lower any price premium, and low productivity will give economic stress to small-scale producers, well-meaning and sincere as most of them are. D T Avery of the Hudson Institute pointed out that data from the Centers for Disease Control in the USA indicated that consumers of organic and 'natural' foods were eight times as likely

to be attacked by the new dangerous strain of *Escherichia coli*, O157:H7. He made the important point that "unless the organic movement puts a voluntary ban on the use of animal manures on food crops, certifies its compost as free of deadly bacteria or irradiates its produce, the health authorities should step in. In the deadliness contest, the bacteria are beating pesticide residues by a score of hundreds to zero". Manures are also associated with protozoan (*e.g. Cryptosporidium*), nematode and other parasitic infections. The UK Soil Association, the organic farming organisation in the UK, bans the use of sewage which in some conventional farming systems is used in the raw and partly treated state.

Organised consumer groups expressed concerns about trade agreements related to the liberalisation of global trade, especially the operations of the WTO, and transatlantic trade between the EU and the USA. Their concerns related to the possibility of lower standards in food and product safety. Of particular interest was the Multilateral Agreement on Investment (MAI) generated by the 29 member countries of the OECD. Consumer groups were concerned about the MAI giving free rein to multinationals to override local and national consumer and environmental regulations. Private industry, profit-generating multinational companies and new technologies were the unifying features of consumer-group attention and ire.

The development of effective consumer policies in the non-Western world was addressed to some extent by meetings and activities in 1998 such as the Euro-Mediterranean Forum on Consumer Policy, the First Regional Conference on Consumers and Public Utilities to address Latin American concerns, the March 15 World Consumer Rights Day on the topic of 'Poverty: Rallying for Change', and the start of the three-year programme by Consumers International on 'Consolidating and Strengthening the Consumer Movement in Africa'.

The Environment

Founded on breathtaking naïvety about 'stabilising' climate change and consequential ecological change by adjusting a few variables, international efforts were made to control atmospheric emissions. There was an inherent ignorance about the reality of dynamic ecosystems, of adaptation, and the need for sophisticated understanding of change. Greater numbers of weather perturbations, many difficult to predict, are anticipated as a result of changing atmospheric composition.

Arising from the UN Framework Convention on Climate Change, held in Kyoto, Japan, in December 1997, representatives of the 160 signatory nations reached the agreement referred to as the Kyoto Protocol to reduce global atmospheric emissions by about 5.2% by 2012. Binding commitments were not demanded of the LDCs. A Clean Development Mechanism was devised to give MDCs incentives to introduce emission-reducing technologies to LDCs. The EU agreed to reduce emissions by an average of 8% below 1990 levels, followed by the US (7%), and Japan (6%); 21 other industrial countries offered to reduce emissions. Prior to the meeting, the World Bank prepared the Global Carbon Initiative to allow developed countries to finance energy-efficient schemes in LDCs. Any savings in 'greenhouse-gas' emissions could then be credited to the binding emissions target of the donor countries under the 'joint implementation' system. Possible serious economic repercussions have delayed the US throughout 1998 from ratifying the proposal, given that the US accounts for 20-25% of global emissions. The USA consumed about 12,3000 kWh *per capita* in 1994 compared with about 1,000-1,500 in China, Thailand and Brazil. Various groups discussed the setting up of national and international 'emissions trading' schemes to help nations meet legally binding commitment to reduce their emissions of greenhouse gases. In November 1998, the independent International Emissions Trading Association held its inaugural meeting under UN auspices.

Although energy efficiency and renewable energy are the most straightforward routes of cutting greenhouse-gas emission, other technologies based on abstracting carbon dioxide from the atmosphere may be viable in the longer term. These technologies include storage of the abstracted carbon dioxide in (i) oceans, to be trapped in sediments, hydrates or iron-fertilised algae; (ii) terrestrial plants, especially in co-ordination with modern agronomy and forestry that reduces soil carbon oxidation and enhances carbon-trapping soil texture; (iii) geologic formations that can sequester carbon dioxide, such as oil and gas reservoirs and mines; (iv) micro-organisms that can also utilise wastes to produce valuable end-products; (v) chemical conversions to inert materials. (It was estimated that the global 1990 output of carbon dioxide could be contained in a space 10km by 10km by 150m if incorporated into magnesium carbonate); and (vi) construction cement to improve its physical qualities. The total terrestrial biomass was calculated by D

Howell and R G Thomas in 1996 to sequester 827 billion tonnes of carbon, the bulk by tropical rainforests (340 billion tonnes), followed by tropical seasonal forests (120), boreal forests (108), temperate deciduous vegetation (95), temperate evergreen vegetation (80), savannah (27), woodland and shrub land (22), swamp and marsh (14), and temperate grasslands (6).

In April 1998, managers of the Global Environment Facility (GEF) reviewed their support of projects in LDCs, and continued to make financial allocations to projects on climate change (40%), biodiversity (40%), ozone depletion (10%) and water supplies (10%). Greater access to GEF activities was acknowledged to be necessary. Funding was increased to $2.75 billion over three years.

Release of the *Living Planet Report* by the World Wide Fund for Nature, the New Economics Foundation and the World Conservation Monitoring Centre, allowed a comparison of present-day anthropogenic activities on the global environment with those recorded in 1960. Since 1960, the use of freshwater had doubled, carbon dioxide emissions had doubled, consumption of sea fish had more than doubled, and the consumption of wood and paper had increased by two thirds.

In January 1998, the Madrid Protocol to the Antarctic Treaty eventually came into force following ratification by Japan, the last of the 26 consultative (voting) signatories. The protocol, which had been drafted in 1991, strengthened the original 1951 Antarctic Treaty. Travellers, for whatever reason, to the region south of latitude 60^0 must seek permission and submit an environmental impact assessment. Environmental clean-ups started, and mining was banned for 50 years. The next meeting of the Antarctic Treaty nations will be held in Peru in 1999.

The strong El Niño detected in 1997, the strongest since 1982-1983, continued into the first part of 1998 and then faded abruptly. It contributed to heavy winter rains in the USA, drought in Central America and Mexico, and floods and drought in South America. Agriculture in those areas was therefore adversely affected. Above-normal ocean temperatures and sea levels were recorded. Preliminary data from terrestrial and ocean temperature observations world-wide indicated that 1998 would be the warmest year on record. According to the World Meteorological Organisation (WMO), the Earth's average temperature in 1998 was the highest since 1860 (when acceptable recordings were introduced) at nearly 0.6°C above the recent long-term average, and the twentieth consecutive year of above-normal global temperatures. Even so, La Niña, a cold episode, developed in the last six months of 1998. The sea-surface temperature in the eastern equatorial Pacific was several degrees below the long-term mean. As a result, climate-related issues dominated marine and coastal resource management during 1998.

Forest fires were a prominent feature of air pollution events in 1998. Haze attributable to forest and other fires on Borneo blocked sunlight and caused transport difficulties and affected tourism in the region. The Pollutant Standard Index level reached 500 in Borneo, when a value of 300 is regarded as 'hazardous' and above 400 is 'very hazardous'. Central America, especially Mexico, suffered large-scale fires, induced by El Niño.

Attempts were made by several countries to combat air pollution. According to a report published in January 1998 by the UK Department of Health, traffic fumes containing ozone, particulate matter and sulfur dioxide were causing the premature deaths of 12,000-24,000 people a year and causing 14,000-24,000 to be admitted to hospital. EU environment ministers agreed in June 1998 that from 1 January 2000, emissions from petroleum-spirit-powered cars would be reduced by 30-40% and from diesel-engined cars by 50%. The sulfur content of petroleum spirit would be reduced by 70% and of diesel by 30%. These new emission limits would be reduced by an additional 50% from 1 January 2005. China announced its intention to address the massive air pollution problem in 29 of its cities. According to the World Resources Institute in its *Urban Air Pollution Risks to Children: A Global Environmental Health Indicator*, an Environmental Health Indicator has been devised which shows that some of the highest air pollution risks to children occur in cities in Brazil, China, India, Iran, and Mexico. WHO has found that fine particulate pollution is responsible for 7-10% of respiratory infections in European children (21% in the most polluted cities). Urban planting of trees and shrubs can reduce considerably particulate air pollution.

A report by the WMO and the UN Environment Programme (UNEP), published in June 1998, stated that the 1987 Montreal Protocol was actually working. Full recovery of the ozone layer was expected by

the middle of the 21st century but detectable signs of recovery would not be apparent until about 2020.

In 1998, the ozone 'hole' covering the Antarctic was the largest ever recorded, extending over an area of about twice the size of the continent (*i.e.* a 28 million sq km 'hole') and extending higher above the Earth's surface than had been previously measured. A winter chill in the atmosphere leading to increased cloud formation, as opposed to increased levels of anthropogenic chlorofluorocarbons (CFCs) and halons, was given as a possible cause for the extended hole. It is on the surfaces of clouds (aerosols and ice crystals) that the CFCs and halons destroy ozone. There were alarming indications that the West Antarctic Ice Sheet, which if melted would raise sea level by 5.5m, had melted at least once in the last 1.3 million years, and that the Pine Island Glacier was retreating inland by more than 1 km a year. In the Arctic, satellite measurements showed that the southernmost edge of the pack ice near Alaska retreated hundreds of kilometres closer to the pole between 1996 and 1998.

Excess nitrogen in the biosphere caused by the overuse of nitrogen fertilizer, the emission of nitrogen oxides by vehicles and factories, livestock and human excreta, and inadequate terrestrial vegetation cover to act as nitrogen reservoirs, has become a serious problem affecting land use and coastal and estuarine waters. The potential of the natural-abundance stable-isotope facilities and expertise at SCRI to address not only the rôle of nitrogen in the biosphere but also policy matters relating to pollution has been appreciated worldwide.

UNEP sponsored the fourth conference of Parties to the Basel Convention on Waste Management in February 1998 in Malaysia, and sought support for the 1995 ban on the export of toxic waste from industrialised to industrialising countries. The meeting agreed on the content of the list of materials defined as hazardous and on a list of countries that were permitted to trade amongst themselves in toxic wastes. Later in the year, in Montreal, UNEP brought together delegates from more than 100 countries to reduce or ban the use of the 12 most dangerous substances, namely, aldrin, chlordane, DDT, dieldrin, dioxins, endrin, chlorinated furans, heptachlor, hexachlorobenzene, mirex and toxaphene. In September 1999, more than 370 scientists appealed to diplomats at a meeting of UNEP not to ban DDT, claiming that it would cost hundreds of thousands of lives in tropical countries where DDT is the most effective way of controlling malarial-vectoring mosquitoes. According to the Malaria Foundation International, malaria kills 2.7 million people a year.

The 1998 edition of the UN List of Protected Areas described a global network of more than 30,000 protected areas totalling 13.2 million sq km designated under national legislation of varying levels of rigour to conserve nature and associated cultural resources. One of the world's largest and most undisturbed tropical forests was permanently protected in June 1998 when Suriname created a 16,200 sq km reserve covering 10% of its land area. Fragmentation of habitats leads to 'island biogeography' symptoms of ecological change (*viz*. a disproportionate decline in the numbers of species, inbreeding, vulnerability to biotic and abiotic stresses, and the edges or margins of the fragments become vulnerable to damage).

Habitat reconstruction (ecological restoration) to restore damaged lands and waters is a special challenge for mankind. Benign neglect, specific plantings, removal of sources of contamination, elimination of 'alien' species, prevention of erosion, introduction of natural disturbances (*e.g.* controlled flooding or burning) were all attempted in 1998. Success, however, is dependent on a full appreciation of biodiversity of the flora, fauna and soil; its measurement and conservation; and the scale of the area to be restored. According to a report in 1998 from the World Wide Fund for Nature (http://www.wwf-uk.org), 154 animal and plant species became extinct in the UK. The current rate of species extinction was calculated to be three species every 2 years. Since the last century, 95% of Britain's peatlands, 95% of its original grazing marshland and 50% of farm ponds have been lost. During the last 50 years, 50% of the ancient semi-natural woodland has gone, and between 1947-1985, 175,000 kilometres of hedges in England and Wales have been lost.

More than 400 delegates from 55 countries attended the fifth International Botanic Gardens Conservation Congress in Cape Town, South Africa, where a two-year review process was launched by Botanic Gardens Conservation International to consider the international Botanic Gardens Conservation Strategy. At a time when the Convention in Biological Diversity should be implemented, botanic garden and research institutes world-wide were mindful of their responsibilities even though the care and maintenance of *in situ* and *ex situ* collections were under financial strictures. The World Conservation Union published that

nearly 34,000 of an estimated 270,000 plant species were under threat. From a 3-year study, *The World of Threatened Trees*, compiled by the World Conservation Monitoring Centre, it was concluded that more than 10% of the world's estimated 80,000 to 100,000 tree species are at risk. In Britain, 11 *Sorbus* species were deemed to be at risk with *S. leyana* (Ley's whitebeam), *S. wilmottiana*, and *S. leptophylla* (Welsh whitebeam) classified as critically endangered. Funding was starting to be released on a small scale for the conservation and exploitation of medicinal plants, using public and private funding sources, partly to respond to interest in alternative medical treatments based on herbal products.

At a meeting of environmental ministers for Canada, France, Germany, Italy, Japan, Russia, the UK and the USA held in April 1998, it was stated that the trade in illicit drugs was the only illegal industry that generated more money than the $5 billion a year produced by the trade in endangered and rare species.

Agriculturally relevant genetic resources continued to represent a key feature of the CGIAR in concert with FAO. The CGIAR held in trust for FAO a total of around 600,000 accessions of the major food crop, forestry and agro-forestry species in its genebanks, and through the International Plant Genetic Resources Institute helped co-ordinate genetic-resources activities globally. SCRI sustains several important gene banks. The two major types of conservation, *viz. in situ* (*i.e.* maintenance of genetic diversity in its original environment where it is still a functional element of natural or agricultural ecosystems from which it acquires its particular characteristics) and *ex situ* (*i.e.* collections not in their original environment and which may be held in seed or propagule form, *in vitro* tissue culture, cryopreservation, pollen banks, seed/clonal orchards, and potentially in DNA libraries) were of special concern to the CGIAR. In 1993, the Convention on Biological Diversity recognised the genetic resources occurring in a country as the sovereign property of that country, as stated in Article 15(1). It also incorporated, for the first time in international agreements, the principles of ethics and equity in both access to genetic wealth and sharing of benefits. The concepts of 'biopiracy', *i.e.* not paying for source genetic material, 'bioprospecting' *i.e.* seeking out new or novel genetic material and compounds derived from it, and 'green imperialism' *i.e.* imposing conditions on LDCs in respect of ecological exploitation, were frequent topics of debate.

Seed crops for domestic and commercial horticulture were affected in 1998 by adverse weather conditions in Africa, Europe and the USA. Around one third of the bulb crop in The Netherlands was lost. The popularity of gardening in the Western economies was both stimulated and exploited by the press, broadcast media and the web, strongly influencing purchasing patterns regionally and nationally for both plants and gardening sundries. Few practitioners seemed to realise the gene-flow implications in gardening of the perpetual practice of introducing novel and alien species (exotics), with their actual as well as potential pests and diseases, whilst many high-profile gardening practitioners in the media openly deprecated the introduction of GM crops in agriculture.

United Kingdom Perspectives

Primacy of place in the list of key events at the end of the millennium affecting the future of the UK was constitutional change – devolution, and fundamental change to the unelected House of Lords by eliminating, or reducing to a rump, participation of those who have hereditary rights of membership (those that remain, hereditary or otherwise, would apparently and anachronistically retain the privilege of titles). No clear rôle or system of election was mapped out for the House of Lords, a body that was noted for its important reports on scientific and associated matters.

Devolution arrangements with the accompanying constitutional changes, proceeded rapidly in the UK following the elections for the Northern Ireland Assembly. Elections for the Scottish Parliament took place in May 1999, and the responsibilities of the Scottish Office Agriculture, Environment and Fisheries Department (SOAEFD) that sponsors the five Scottish Agricultural and Biological Research Institutes including SCRI, were largely devolved to the Scottish Executive Rural Affairs Department (SERAD). Elections also took place in May 1999 for the National Assembly of Wales, with the Assembly largely inheriting the Welsh Secretary of State's agricultural responsibilities. The new Scottish Parliament comprising 73 constituency members and 56 regional

members assumed its full legislative powers on 1st July 1999. The 'Barnett formula' will still be applied to calculate the 'Scottish Block' (approximately $15 billion) for funding together with payments from other sources to comprise the Scottish Consolidated Fund. It has been claimed that the Barnett formula has given Scotland an overly generous financial settlement, a fiscal subsidy in the order of £3.9 billion according to Chantry Vellacott DFK, and there may be pressure to reform the basis of calculation.

Of greater immediacy to the public, however, was the economy which started 1998 in a state of relative buoyancy, capitalising on the previous 5 years when annual growth in output exceeded the long-term trend of 2.25%. Adoption of common EU statistical practices meant that the national accounts were rebased, and revision to historic economic indicators showed that the annual average increase in real GDP since 1991 was 0.25 percentage point higher than previously calculated. As 1998 progressed, however, the economy lost a certain amount of momentum, and by the end of 1998, business confidence had fallen and there was even discussion of a short period of recession, but fortunately that failed to materialise in 1999. Economic growth was sustained by the domestic economy and business investment rather than the trade sector. The dominant service sector, accounting for 60% of output, outperformed the rest of the UK economy, particularly in the telecommunications and transport segments of the market. Although only accounting for 20% of output, the manufacturing sector was a major consumer of services. Manufacturers and exporters suffered from the strength of the pound. Despite weak official retail sales data, consumer spending was clearly resilient, indicative of the fact that the purchase of goods and services was increasingly outwith the recording of official statistics. There is a need to portray accurate consumer spending and inflation data to avoid serious economic policy mistakes. External factors influenced the rate of inflation, rather than the actions of the Bank of England's Monetary Policy Committee (MPC). Charged with managing interest rates to facilitate an economic growth rate commensurate with low inflation, the MPC was set a benchmark of 2.25%, growth above which was rather simplistically considered inflationary. Given the need to achieve multiple economic objectives, some economists were concerned that monetary policy targeted on inflation, and fiscal policy focused on budget consolidation, would exclude unnecessarily other valuable policy instruments. Consumer prices in 1998 were expected to have risen by 2.7% in 1998, similar to 1997, but the impact of taxes, seasonal changes in food prices, retail sales practices, commodity prices and the costs of imported goods had yet to be assessed. The costs of services rose by about 5% a year in 1998. Average earnings growth was constrained in the public and manufacturing sectors to an overall figure of 4%, but this was expected to decline in 1999 as a response to falling corporate profits. Job creation was healthy as the number of employed exceeded 27 million for the first time. At 6.2%, unemployment was at its lowest since 1980.

The UK Government moved to adopt measures on low pay and union recognition. In May, the Commission on Low Pay recommended that there should be a natural minimum wage, the Government favouring a rate of £3.60 per hour for workers 22 years old and over, with a rate of £3.00 an hour for workers aged 18-22, subsequently increased to £3.20. In the same month, the Government published its proposals on employment rights in the White Paper *Fairness at Work*, obliging employers to recognise trades unions in cases when at least 40% of eligible employees voted in favour of having a union, with automatic recognition when more than half of the relevant workforce belonged to a union. For claims alleging unfair dismissal, the qualifying length of service would be reduced from 2 years to 1 year. A right for time off for urgent family reasons would be introduced, and statutory maternity leave would be increased from 14 to 18 weeks.

Political lobbying started to bring about a reduction in the impact of proposed energy and pesticides taxes. According to the National Farmers Union of England and Wales, agriculture and horticulture would suffer an energy tax of about £26 million a year, offset by national insurance rebates of about £9 million a year, and the glasshouse sector would be hit with extra costs of about £12,500 *per* hectare. Other EU member states were thought to have designed their tax regimes such that the impacts of these taxes on agriculture and horticulture would be marginal. According to the Centre for European Agricultural Studies, a pesticide tax alone would abstract up to £320 million from agricultural and horticultural incomes. Livestock-based agricultural enterprise faced actual and impending charges imposed by the Meat Hygiene Service, welfare controls, the Cattle Tracing Scheme, and assurance and other traceability schemes needed to restore the credibility of the meat industry in the post-Bovine-Spongiform-Encephalopathy, and post-food-

poisoning era. By the end of March 1999, figures from the EC indicated that the number of cattle recorded as suffering from BSE were 176,326 in the UK, 373 in the Irish Republic, 293 in Switzerland, 250 in Portugal, 57 in France, 8 in Belgium and 6 in Germany and also in The Netherlands. Alarmingly, A Charon, Director of the veterinary service in the Mayenne Department of France, stated that his inspectors had checked just 28 out of 700,000 cows for BSE this decade. According to Reading University in *Geoforum*, the BSE crisis will lead to the loss of more than 44,000 jobs in England alone. Many agricultural producers throughout 1998-1999 faced catastrophic losses, affected by oversupply and the strength of the pound sterling.

Sole traders and partnerships, a common feature of agriculture and horticulture, were being edged towards incorporation, according to NatWest. The cost of compliance with current tax rules cost small businesses, both companies and unincorporated, between £5-17 billion a year, with compliance costs falling most heavily on the smallest firms. Red-tape costs represented 18% of the total tax burden of sole traders and partnerships, but only 4% of limited companies. The biggest problems for small businesses were value-added tax, PAYE and National Insurance taxes, and tax self-assessment.

A study by Manchester University noted that inspectors have the right of entry to business premises under 267 different statutes. In 1998, more than 24,000 staff were associated with the official inspection of business, making 466,000 inspection visits. There were nearly 10,000 local authority staff responsible for enforcing legislation, nearly 6,500 of which dealt with health and safety. In 1979, there were 116 fewer statutes giving inspectors the right of entry and search. Although EU-sponsored inspectors have the right not only to conduct investigations regarding competition law on business premises, but also seek information from the suspect firm's associates, suppliers and customers, the intrusion must be 'necessary' and not 'arbitrary or excessive'. Unfortunately, there is no guidance or case law on the meaning of these words. The major problems relate to the excessive number of rules; the speed, volume and complexity of law-making; inconsistent enforcement across the EU; the adversarial attitudes of the inspectors; and crucially, the costs of compliance. A Fair Regulation Campaign has been established, involving a wide range of organisations in industry and commerce.

From an analysis published by the Department of Trade and Industry in *The UK R&D Scoreboard 1999* of 561 UK and 300 international R&D investing companies, there were eight points of significance (see http://www.innovation.gov.uk/finance). (i) On average, UK companies increased their R&D spend since last year by 6% compared with a 12% increase by the 300 international companies. (ii) The international companies included 16 UK companies which account for more than 50% of the R&D invested by the 561 companies in the UK list. (iii) The aggregate R&D intensity (R&D/sales) of the top 16 UK companies was 2.9% compared with 3.1% expected if these companies had invested at the same intensity as the international rate for its sector. (iv) The aggregate intensity of the 300 international companies was 4.9%. (v) The UK pharmaceutical sector continued to lead the world with 15% of sales invested in R&D as against 13.5% internationally. (vi) Overall, the 561 UK companies had comparable profitability (profits/sales) of 11% with the 10% level of international companies. (vii) Around 75% of UK companies which invest in R&D at more than 2% of sales showed a commitment to innovation by increasing their R&D even when their profits were falling. (viii) The Scoreboard did not include UK companies which undertake R&D but did not declare the amount in their accounts.

It is axiomatic that competitiveness, especially in the medium to long-term, requires innovation and investment in R&D as a key economic driver, and it is therefore pleasing that the Scoreboard data showed that many UK companies invested on a par with international competitors. Specific circumstances determine the extent to which a stream of new products, processes and services are required in the future. Moreover, the effectiveness of the R&D effort is critical, bringing together management skills, marketing, capability, finance, and legal protection. According to E George of the Bank of England, there were 381 high-technology firms listed on UK markets, with a combined market value of £1,281 billion and making up 30% by value of the London market. Over the course of 1998, the value of the Financial Times Stock Exchange index increased by 5.5%, but the index of UK high-technology companies increased by 89%.

With regard to R&D intensity (R&D/sales) and profitability (profits/sales), five technology-based sectors were chosen for analysis in the *R&D Scoreboard 1999*. In the chemicals sector, nearly all the UK companies

had a lower R&D intensity than their foreign competitors and their aggregate profitability was lower at 8.1% compared with 9.6%. In the pharmaceutical sector, the UK companies had a higher aggregate R&D intensity than their foreign rivals but their aggregate profitability was lower at 21.7% compared with 23.9%. In the engineering and machinery sector, the picture was mixed with a few companies investing heavily but most with low R&D intensities: the aggregate R&D intensity of UK companies was at 1% compared with 3.3% for international companies, but aggregate profitability was higher at 10% compared with 7.6%. For the electronic and electrical sector, the picture was similarly mixed with only a few heavily investing companies matching their overseas rivals; aggregate R&D intensity was 3.2% for UK companies and 5.3% for international companies, and aggregate profitability was 6.1% compared with 10.6%. Finally, the international software and information technology Sector revealed that the impact of the UK companies was focused mainly on niche markets. Aggregate R&D intensity was 4.9% for UK companies and 13.6% for international companies; aggregate profitability was 6.9% for the UK companies, vastly less than the 24.2% for international companies.

Analysis of R&D intensity values in the G5 countries (France, Germany, Japan, UK and USA) revealed that the UK value was substantially less than those of the other nations.

During recent years, there have been numerous attempts to emulate the entrepreneurial culture of the USA. In the USA, the enterprise culture is incalcated into the framework of society with more than one in 12 people, or a third of households, having a family member who has started a small business. The US tax system and public attitudes encourage risk-taking, business regulation albeit tough is flexible, and support services for new firms (lawyers, accountants, estate agents and planners) are perceived as helpers rather than hinderers. In preparing to launch the Small Business Service, based loosely on the highly effective American Small Business Administration, the UK Government planned for the new service to act as a voice for small firms, to simplify and improve governmental support, and monitor regulations. According to The Global Entrepreneurial Monitor, only 16% of people in the UK thought there were good opportunities to start a business in the near future compared with 57% in the USA. Attitudes to the status of entrepreneurs, too, were much more negative in the UK than in the rest of the world, and there was evidence that the UK was losing its entrepreneurial edge. Counteracting entrepreneurial behaviour in the UK, however, was the increasing bureaucratic burden placed on industry. Small businesses will not be exempt from administering the new stakeholder pension. Together with the newly imposed working time directive, the national minimum wage, the fairness at work proposals, and the administration of the new working families tax credit, it was estimated that the cumulative effect of such measures will add £4.6 billion to business costs and ultimately eradicate 880,000 jobs, according to P Minford and A Haldenby in *The Price of Fairness*.

Further evidence of the UK's poor showing as a generator of wealth was presented in an analysis in December 1998 by Professor M Porter of the Harvard Business School. The UK's ranking in an 'innovation index', based on patents, R&D spending and outputs, and ability to derive commercial benefits from science and technology was 13th out of 17 MDCs, and is likely to slip to 15th by 2006. The 1996 innovation index ranking was in the following order: USA, Japan, Switzerland, Germany, Denmark, Sweden, France, Canada, Finland, Norway, The Netherlands, Australia, UK, Austria, Italy, New Zealand, and Spain. Attitudes to entrepreneurs becoming 'too rich', capital gains taxation, difficulties of defraying R&D costs, accessing venture capital, and crassly inept implementation of government policies were seen as strictures by many independent observers of the UK scene. When international patents are linked with science citations, the UK's figure for 1996 of 87 patents per 1,000 citations puts it in 12th position among 17 countries, below Japan (488), Germany (267), USA (266), Switzerland (266), Austria (150), France (135), Sweden (130), The Netherlands (120), Italy (108), Canada (97), and Finland (89), but above Denmark (57), Norway (56), Australia (46), New Zealand (27) and Spain (25).

In *Driving Productivity and Growth in the UK Economy*, published by the McKinsey Global Institute in 1998, a persuasive case was made for a skilled labour force, a high level of investment, and a stable economy being consequences, as much as causes, of a nation's productivity performance. Lack of competition, excessive regulation, and restrictions on land use were seen to be factors causing the productivity gap between the UK and USA, but surely did not account for the gap between the UK and such heavily regulated countries as Germany and Japan.

Provisional data produced in the excellent reports by the Ministry of Agriculture, Fisheries and Food (MAFF; see http://www.maff.gov.uk/; *Agriculture in the United Kingdom* – produced by MAFF, SOAEFD, Department of Agriculture for Northern Ireland, and the Welsh Office; *Basic Horticultural Statistics for the United Kingdom – Calendar and Crop Years 1987-1997*) indicated that the contribution of agriculture to GDP, using current prices, declined from 1.2% in 1997 to 1.0% in 1998 and followed a long-term trend of decline; in the period 1987-1989, the average contribution was 1.6%. When subsidies were taken into account, the net value of UK agriculture was approximately 0.4% of GDP, an historic and worrying low. About 2.3% of the UK workforce was employed directly in agriculture, a figure that omits many groups whose employment is dependent on primary production such as many employees in the public sector, food processing and industrial feedstock industries. Importation of food, feed and drink amounted to £17,114 million, representing 9% of total UK imports; imports of alcoholic drinks amounted to £2,479 million. A word of caution is needed, however. In recent years, a higher proportion of processed as opposed to unprocessed commodities has been imported. Statistical factors have been introduced to devalue processed imports to the value of their unprocessed food content. This has reduced the estimated value of food imports, which in turn reduced the estimated value of food consumption, and thus UK food production as a percentage of UK food consumption has increased. Exports of food, feed and drink declined from £9,924 million in 1997 to £9,185 million in 1998, of which alcoholic drinks contributed a phenomenal £2,720 million. These agriculturally related exports amounted to 5.6% of total UK exports, down from 5.8% in 1997 and an average of 6.3% in the period 1987-1989. The UK was 68.1% self-sufficient in all food types in 1998, compared with 68.7% the year before, and an average of 72.8% in the period 1987-1989. For indigenous-type food, however, the UK was 82.3% self-sufficient. Household final consumption expenditure on household food and alcoholic drinks at current prices was up from £82,312 million in 1997 to £83,400 million in 1998; astoundingly £29,600 million expenditure was for alcoholic drinks. Household food and alcoholic drinks accounted for 15.9% of total household final consumption expenditure in 1998, down from 16.5% in 1997. Compared with an average of 13.0% expenditure in 1987-1989, household food accounted for just 10.3% of expenditure in 1998.

According to *UK Snacks 1999*, produced by Datamonitor, the UK market for snack foods (crisps, nuts, tortilla chips, cereal bars, extruded savoury snacks, exotic 'nibbles' *etc*.) reached £2.25 billion in 1998. This sector of the food industry recorded a growth rate of about 6.5% *per annum*. Increasing leisure time and the trend towards 'grazing' (eating less but more often, frequently outwith the home) at the expense of the traditional family meal, pointed towards further expansion of the market. The only part of the market that was recorded as declining was nuts, especially peanuts, and crisp sales were static, but sales of cereal bars and extruded savoury snacks were buoyant.

In June 1998, the total area of agricultural land, including common rough grazing, was 18,593,000 hectares, of which 4,972,000 hectares were devoted to crops, and 34,000 hectares were left fallow. In the period 1987-1989, an average of 18,974,000 hectares were committed to agriculture, 5,223,000 hectares of which were harvested for crops. More detailed analysis of the cropping data reveals that the area devoted to cereals declined from 3,514,000 hectares in 1997 to 3,420,000 hectares in 1998, mainly attributable to a decline in the barley area from 1,359,000 to 1,255,000 hectares. The potato area, affected by wet weather in the north and west of the UK which impeded harvesting, declined from 166,000 hectares in 1997 to 164,000 hectares in 1998. Other arable crops, excluding potatoes, were grown on an increased area of land, up from 1,126,000 hectares in 1997 to 1,210,000 hectares in 1998. This increase was accounted for by enlargement of the areas cropped for oilseed rape, up from 445,000 hectares to 506,000 hectares, and field beans, up from 197,000 hectares to 213,000 hectares. The area for sugar beet fell from 196,000 hectares in 1997 to 189,000 hectares, and the area of land for horticulture, too, declined, from 11,633 hectares in 1997 to 11,519 hectares in 1998, a drop of around 4%.

Without taking account of direct subsidy payments, the average price of agricultural products fell by 9% between 1997 and 1998, and inputs fell by 3%. The average price of agricultural products was 5% lower than 10 years previously whereas the average price of inputs increased by 28%. The value of the output of all agricultural commodities fell by £1.7 billion or 9%.

In terms of production, cereals declined by 800,000 tonnes to 22,692,000 tonnes in 1998, from

23,533,000 tonnes in 1997. The value of production also declined from £2.913 million to £2.493 million. Cereal yields in 1998 were 7.56 tonnes per hectare for wheat, 5.29 for barley, 6.00 for oats, 4.88 for triticale. Wheat production increased from 15,018,000 tonnes in 1997 to 15,449,000 tonnes valued at £1,647 million in 1998. Barley, one of SCRI's mandate crops, declined over the same period from 7,828,000 tonnes to 6,537,000 tonnes valued at £777 million. Oat production increased from 577,000 tonnes to 588,000 tonnes valued at £59 million.

Potato production in 1998 sharply declined from 7,125,000 tonnes in 1997 to 6,505,000 tonnes in 1998 valued at £639 million. In 1995, the crop which is a key mandate crop for SCRI, was valued at £1,077 million. Oilseed rape production increased from 1,527,000 tonnes in 1997 to 1,569,000 tonnes in 1998 valued at £399 million. Sugarbeet production in 1998 was estimated to be 9,802,000 tonnes, adjusted at standard 16% sugar content, and was valued at £274 million, the lowest for several years. Since 1987-1989, the area of land put down to linseed has increased from 13,000 hectares to 101,000 hectares in 1998. The low yield of 1.41 tonnes *per* hectare, gave a volume of 143,000 tonnes in 1998, valued at £67 million.

Horticultural production was valued at £1,903 million in 1998. Vegetables grown in the open on an area of 160,400 hectares were valued at £667 million, and £298 million for protected crops on an area of 1,400 hectares. The highest valued horticultural commodities were mushrooms (£168 million) and carrots (£104 million), followed by lettuces (£74 million), peas (£58 million), tomatoes (£57 million), cabbages (£51 million) and cauliflowers (£43 million). Orchard (top) fruit production on an area of 21,900 hectares was valued at £86 million, and soft fruit at £111 million on 10,100 hectares, mainly attributable to two crops of special importance to SCRI, strawberries and raspberries. Ornamental production on 19,700 hectares was valued at £658 million, attributed to £332 million for hardy ornamental nursery stock, £284 million for protected crops and £42 million for flowers and bulbs in the open. MAFF estimated that the measure 'Total Income From Farming', which is sensitive to small changes in the values of outputs and inputs, was £2.2 billion in 1998, some 29% or £900 million less than in 1997. Income to paid workers directly employed in primary-production agriculture was estimated to have been £1.9 billion, 2% more than in 1997. Net farm income, deflated by the retail price index (*i.e.* in real terms as opposed to current prices) and using indices whereby 1989/1990 to 1991/1992 = 100, were just 50 for cereals, 70 for general cropping and –20 for mixed farming; the figure was –25 for cattle and sheep in the less-favoured areas.

The total UK public expenditure on agriculture in 1998/1999 was forecast to decrease by £28 million from the previous year to £3.555 billion. Spending under CAP was forecast to decrease from £3.321 billion to £3.293 billion, of which 35% was devoted to the arable areas payments scheme, 5% to cereals and 5% to sugar. Policy developments in the arable sector during 1998/1999 included the retention of the Home Grown Cereals Authority (an important statutory levy board relevant to SCRI), revision of the agri-monetary system, derogation on the moisture content of grain offered to intervention, a reduction in area payments specific to oilseeds, increase in the level of obligatory set-aside to 10% for the 1999 harvest, changes to the EU implementation rules for the reformed fruit and vegetables regime, tighter limits on the growth of hemp for fibre, assistance to hop growers to lessen oversupply, and a limited area allocation of 305 hectares for grubbing up apple and pear orchards.

The Sugar Beet Research and Education Committee was wound up at the end of March 1999 and the system of funding sugar-beet research by means of a statutory levy would be terminated as from April 2000. Payments were made covering management agreements and conservation plans under the Environmentally Sensitive Area Scheme, the Countryside Stewardship Scheme, the budget-busting Organic Aid Scheme for farmers converting to organic production, the Habitat Scheme, the Farm Woodland Premium Scheme, the Countryside Premium Scheme in Scotland, Tir Gofal (Land Care) starting in April 1994 in Wales, Nitrate Sensitive Areas Scheme and Nitrate Vulnerable Zones. There were also subsidies paid to farmers to support capital improvements and to compensate for the loss of capital assets.

In order to make the UK system of plant breeders' rights compatible with the 1991 Revisions to the International Convention for the Protection of New Varieties of Plants, the Plant Varieties Act 1997, with its subordinate legislation, came into force in May 1998. Amendments were made to the Seeds (National Lists of Varieties) Regulations 1982 to remove the redundant requirement for applications to be accompanied by trial data from the applicant.

From now on, the addition of varieties (cultivars) to the National List will be made solely on the basis of official trials.

Land reform plans in Scotland, giving increased public access not only to remote, wild areas but also to low-ground arable and stock farmland, raised concerns in the farming community. In addition to potential adverse effects on conservation, there were worries about privacy, safety, and compliance with legal and quality-assurance standards jeopardised by undesirable effects of litter and pets. Another aspect of land reform to permit a local community to register an interest in an area of land or estate, and whilst that interest has been registered, if the land is put up for sale it must be offered to the interest group at a price set by a State valuer, was perceived by many rural landowners as controversial and tantamount to land nationalisation.

Announcements were made to set up an independent and transparent Food Standards Agency with a Scottish counterpart, focusing on protecting consumer health in relation to food, and with powers to act throughout the whole food chain. It was intended that the new Agency would interact with all interested parties to develop policies that are proportionate to the risks involved.

Plant Biotechnology

The OECD defined biotechnology as "the application of scientific and engineering principles to the processing of materials by biological agents". More formally, biotechnology refers to the application of organisms, sub-cellular entities, or biological processes, to manufacturing and service industries, including agriculture, horticulture, forestry, human and veterinary medicine and pharmaceuticals, food production and processing, and environmental management such as bioremediation. The aims of the technology encompass: biomass production; production of chemicals and useful products; purification of water; decomposition of wastes and recovery of valuable components; generation of new types of organisms; exploitation of fermentation; diagnosis, prevention and treatment of diseases; unravelling metabolic pathways; and propagation of cells and whole organisms. Recent technological and intellectual advances in molecular genetics – particularly sequencing of genes and proteins, isolation and insertion of genes into receptor organisms, development of marker genes and promoters, and gene amplification – have given rise to the 'new biotechnology'. The techniques of biotechnology are being used in fundamental research in the life sciences, and the divisions between plant, animal and microbial biotechnology are becoming increasingly irrelevant.

New interfaces within biotechnology include those with information technology (bioinformatics), chemistry (new separatory and identification systems, novel biomaterials), physics and engineering (gene chips), electronics (biomolecular computing and molecular design), and nano-scale engineering and medicine (molecular-scale surgery, tissue engineering) *etc*. A new branch of biotechnology in both the private and public sectors has achieved special prominence – genomics, which covers structural genomics (determination of the complete nucleotide sequence of a genome and identification of its genes) and functional genomics (characterisation of gene function on a genome-wide scale). Most plant genomes are largely unexplored. In view of the fact that genes are studied in groups in parallel, computational analysis of the data, *via* bioinformatics, is central to any genomics strategy, which will concertina the time to bring discoveries and inventions to the marketplace. Biotechnology in all its guises affects all areas of human activity, and no nation can afford to ignore the huge potential of the range of biotechnological developments coming on stream.

In Europe in the last financial year, there were 1178 biotechnology companies employing 45,823 staff. In the USA, there were 1283 biotechnology companies employing 153,000 staff. The areas of activity covered platform technologies, contract R&D and manufacturing, therapeutics, diagnostics, biochemicals, agbiotech, environmental, food processing, and related services. All enjoyed substantial rates of growth and output.

Regardless of attempts throughout 1998-1999 by senior politicians, industry, biotechnologists, molecular geneticists and eminent scientists to convince UK consumers and growers of the benefits and opportunities provided by agricultural biotechnology, there was evidence that public and lower-level political support waned substantially in the face of high-profile anti-biotechnology accounts in the press and on television. This has been attributed to the impacts of BSE and the perceived inadequacies of industry, government and scientists in permitting the situation of contaminated beef to arise. To this we must add the general ignorance of the public and politicians in the understanding of risk (*e.g.* cars, cigarettes, stepladders, playing sports *etc.* are dangerous, eating GM food is not),

and of understanding in general science, engineering and technology. Environmentalist groups monitored the location and progress of GM crop trials, facilitating widespread destruction of crop trials throughout Europe. Ecoterrorists (eco-warriors, eco-vandals) of various affiliations exploited benign governmental attitudes to private property by vandalising trials in France, Germany and the UK; 27 of the 163 trials in the UK in 1998 were vandalised, a trend that continued with increasing media attention in 1999. Organisations that enjoyed the taxation benefits of charitable status were implicated in the illegal attacks.

Pejorative and wholly unjustified language such as 'Frankenstein Foods', 'unwanted', 'contamination', and 'genetic pollution' (see p. 45), was used to stimulate negative emotions in Europe. Rational and detailed debate was rare. Not surprisingly, whilst the area of GM crops expanded in the rest of the world, commercial plantings of transgenic cultivars were constrained in Europe. The UK introduced a 1-year ban on commercial plantings, and a 3-year ban on GM insect-resistant crops. A ministerial committee was proposed to oversee policy developments relating to biotechnology. In August 1998, the French government imposed a 2-year moratorium on the planting of GM oilseed rape, and in December, established a permanent 'biovigilance' committee to examine the safety and environmental impact of GM crops. Also, in France, there were legal appeals against the approval of GM maize lines from three companies, and the authorities did not ratify a GM oilseed rape cultivar that had received EU authorisation in June 1997. The Greek government banned the import and marketing of a glufosinate-tolerant oilseed rape, despite EU authorisation. Similarly, Australia and Luxemburg proved a hostile environment for the trialling of corn-borer-resistant maize; the European Parliament and Commission agreed in December 1998 to postpone attempts to overrule the bans imposed by both countries. In contrast, only 70,000 signatures were collected for a petition to ban all GMOs, and in June 1998 Swiss voters rejected with a majority of 67% a proposal to restrict biotechnology in agriculture and medicine.

In the GM debate, the main concerns were possible (i) risk to human health, (ii) risks to the environment, (iii) regulatory weaknesses, and (iv) ethical unacceptability. With regard to human health, in most countries the safety of traditional foods derived from plants is not regularly reviewed on the basis that foods consumed for generations should be safe as part of a normal diet. Even so, traditional foods contain natural toxins (*e.g.* lectins, glucosinolates, erucic acid, glycoalkaloids *etc*.). There are naturally occurring carcinogens in most plants, as well as carcinogens derived from (a) the frying, baking and smoking of foodstuffs, (b) the products of microbial attacks of fruit, seed and vegetables, and (c) the products of microbial spoilage of stored products. Nutritional and compositional screening tests and knowledge of the parental material will indicate the presence of toxins in the products of both conventional and GM breeding. Likewise, the possibility of allergenicity caused by alien genes can be readily detected.

Risks to the environment as a result of promiscuity by introduced genes leading to unintentional effects on non-target, possibly beneficial organisms, is a complex area. Gene flow at different levels can be detected in all crops, but where there are no native species with which to cross-breed (*e.g*. UK members of the Solanaceae and commercial potatoes), or where the incidence of cross-breeding is extremely rare, then there is no scientific justification whatsoever for assuming that gene flow from GM crops is any different from that which already occurs on a vast scale in conventional agriculture, horticulture, forestry and domestic gardening, and which occurs in natural ecosystems. Nor is it justified at this juncture in assuming that the gene flow is invariably undesirable or harmful. As I have stated in previous *Reports of the Director*, new forms of agronomy involving refugia, dispersal corridors, buffer zones *etc*. will have to be introduced to reassure growers of organic crops, and retard gene-flow effects from introductions of all types. Questions have been raised about herbicide resistance/tolerance genes which might escape from GM crops into the natural flora, especially weeds, which would become difficult to control. Related to this is the use of herbicide on the GM crop leading to toxicological consequences in humans and wildlife, even though there is no evidence for this. As before, proper monitoring and regulatory processes must be in place, and in fact are equally applicable to applications of pesticides, fungicides *etc*. to conventional, organic and GM forms of agriculture, horticulture, forestry, and gardening.

Possible regulatory weaknesses reflect the difficulty of legal systems coming to terms with the rapid progress of science, increasing the vulnerability of legal and political processes to undue influences, protectionism, and unwarranted actions that either stifle or unjustifiably promote any form of technology. An open and

investigative press, expectation of veracity of the spoken and written word from the media, and a professional civil service reinforcing democratic governments are pre-requisites to regenerate public confidence in the stance taken about any proposed technological introduction.

Ethical issues were dealt with in great detail in 1999 by the report *Genetically modified crops : the ethical and social issues* by the Nuffield Council on Bioethics. The view was taken that "the genetic modification of plants does not differ to such an extent from conventional breeding that it is in itself morally objectionable", but "does, however, have the potential to lead to significant changes in farming practices in food production and the environment." All the GM food so far on the UK market was considered to be safe for human consumption. Conventional breeding has itself produced and continues to produce types of crops that do not occur naturally (*e.g. Triticale, Raphanobrassica etc.*) and many crop types incorporate alien genes. Nevertheless, ethical concerns must be respected, not least in scientific reporting. In summary, all four concerns in the GM debate are legitimate, and responses to these concerns must not be shaped solely by industry.

One of the most cited scientists in the international GM debate has been A Pusztai, formerly of the Rowett Research Institute in Aberdeen. By means of television, radio, newspapers and the Internet, rather than subjecting his research to peer review, he claimed in 1998 that diets containing GM potatoes expressing the snowdrop lectin, *Galanthus nivalis* aggutinin (GNA), affected different parts of the rat gastrointestinal tract. Some effects were claimed to be due to the expression of the GNA transgene, but other parts of the construct, or the genetic transformation itself, or both, could also have contributed to the overall biological effects of the GM potatoes. The initial reports were announced as factual in press releases by the then Director of the Institute, W P T James, and the Chairman, J Provan. Shortly afterwards, Professor James launched an audit of the Rowett research on Pusztai's unpublished work. The report of the audit stated "The Audit Committee is of the opinion that the existing data do not support any suggestion that the consumption by rats of transgenic potatoes expressing GNA has an effect on growth, organ development or the immune function. Thus the previous suggestion that the research results demonstrated adverse effects from feeding genetically modified potatoes to rats was unfounded." In April 1999, the Royal Society convened a Working Group to examine whether the publicised but unpublished work would require changes to the Society's September 1998 statement on GM plants for food use. Six reviewers considered the available evidence and concluded that "the safety of GM plants is an important and complex area of scientific research and demands rigorous standards. However, on the basis of the information available to us, it appears that the reported work from the Rowett is flawed in many aspects of design, execution and analysis and that no conclusions should be drawn from it. We found no convincing evidence of adverse effects from GM potatoes. Where the data seemed to show slight differences between rats fed predominantly on GM and on non-GM potatoes, the differences were uninterpretable because of the technical limitations of the experiments and the incorrect use of statistical tests. The work concerned one particular species of animal, when fed with one particular product modified by the insertion of one particular gene by one particular method. However skilfully the experiments were done, it would be unjustifiable to draw from them general conclusions about whether genetically modified foods are harmful to human beings or not. Each GM food must be assessed individually. The whole episode underlines how important it is that research scientists should expose new research results to others able to offer informed criticism before releasing them into the public arena. In view of the public interest in this case we recommend that the results of any future studies on testing GM food safety, when completed, should be peer reviewed and then published. This would provide an opportunity for the international scientific community and the public at large to have access to the information."

In October 1999, S W B Ewan and A Pusztai published their observations and conclusions in a Research Letter to Lancet, accompanied by a pithy commentary from the Editor. Accompanying this commentary was a commentary by H Kuiper and colleagues from Wageningen, and also a Research Letter from B Fenton and K Stanley of SCRI, and S Fenton and C Bolton-Smith of the University of Dundee, Ninewells Hospital and Medical School, on the binding of GNA to human white blood cells *in vitro*. Kuiper and colleagues pointed out that the experiments of Ewan and Pusztai (a) were incomplete, with too few animals *per* diet group, (b) did not report on the composition of the different diets, (c) lacked controls such as a standard rodent diet and a test diet with potatoes containing an 'empty' vector, (d) did not

observe consistent patterns of change, (e) did not deal adequately with possible adaptive changes in the gut because of the low digestability of raw or partly refined starch. Accordingly, Kuiper and colleagues concluded that "the results are difficult to interpret and do not allow the conclusion that the genetic modification of potatoes accounts for adverse effects in animals". They concluded that the work of Fenton and colleagues emphasises the need for further studies on the bioavailability of lectins and potential toxic effects once they have entered the systemic circulation.

The work conducted by Ewan and Pusztai was part of a collaborative study, funded by SERAD, on lectins and their possible use in protecting plants from pest attacks. The GM potatoes were not designed for commercial release into the food chain – the toxicity level of a wide range of lectins is well known and allowed for by avoiding poisonous species or by careful food preparation. Future work will undoubtedly consider expression of certain lectins in non-food crops, or confinement of expression in parts of food plants that are not consumed, or expression for specific periods during development, or expression only at the specific site of pest attack. As Kuiper and colleagues pointed out, unintended effects of genetic modification can be detected by screening for altered metabolism in the GMO by analysis of gene expression (monitored by microarray technology, mRNA fingerprinting *etc.*), by detailed protein analysis (proteomics), and by secondary metabolite profiling (metabolomics). These tests would be in addition to extensive toxicological and nutritional assessments. In truth, such tests could be applied in future to conventionally bred cultivars. What is clear, though, is that all current commercially available GM crops do not differ from the traditionally grown crops except for the inserted traits. As second- and third-generation crops come on stream (see later) then more stringent testing should be introduced. Obvious lessons to be drawn from the GM debate include the requirement for wide consultation with the support of authoritative literature; effective and non-pejorative food labelling and testing of all foodstuffs; research to address gaps in current knowledge; and absolute transparency and open access provided to the public and politicians.

Recent EU legislation relevant to plant biotechnology was noted in six areas. Firstly, Novel Food Regulation 258/97, concerning novel foods and novel food products came into force in May 1997 and was extended (1813/97) to cover the labelling of GM maize and soybeans approved prior to May 1997. Implementation of 1813/97 was due to take place in November 1997 but was delayed because the testing methods and label wording were contested. Consequently, it was replaced by 1139/98 which came into force in September 1998, enforcing the labelling of GM maize and soybeans. In deciding to label all products containing modified DNA or proteins, the European Commission has yet to establish a list of products that do not need to be labelled, or even propose threshold levels to be set for GM ingredients in food. The Commission was also working on a draft document to extend GM labelling rules to include additives and flavourings. Secondly, the new Directive (98/81) revising the contained use of GMOs Directive (90/219) came into force in December 1998. Thirdly, the Biotechnology Patents Directive (98/44) harmonising EU rules on the legal protection of biotechnology inventions, was adopted and published in the Official Journal in July 1998, and is due to come into force in July 2000. Fourthly, the European Commission proposed to revise the deliberate release of GMOs Directive 90/220 to update rules on the marketing of food, feed and seed containing or derived from genetically modified material in order to take into account new policy on labelling and scientific evaluation. This proposal related to Directive 79/35, which adopted 90/220 to make labelling compulsory for produce containing GMOs and itself was published in the Official Journal in July 1997. Once the amended GMO Registration Directive (90/220) has been adopted, EU member states will be able to adopt or reject European Commission proposals on GMOs by qualified majority instead of a unanimous decision. Fifthly, there was a proposed Directive on seed, including GM seed, with the aim of harmonising registration and labelling of GM seed and to bring the processes in line with novel food legislation. Lastly, in March 1998, the European Commission proposed excluding GM crops and food products from the organic farming classification.

Diversity of the current risk assessment methods and the regulatory framework in EU member states confounded somewhat those attempts in 1999 to streamline environmental risk assessments required by industry groups. The subjective, largely qualitative assessments can be prone to political and perceptual bias. The Forward Studies Unit of the European Commission recommended the establishment of a multinational export forum to establish common methods for environmental risk assessments. At the

beginning of 1999, a House of Lords Select Committee commented on the regulation and risk assessment of GMOs. The Committee recommended continued risk assessments of GM crops to include delayed and indirect effects, clarification of labelling rules for GM ingredients in food, and revision of Directive 90/220. Biotechnology companies were encouraged to develop alternatives to antibiotic resistance markers in GM crops.

Elsewhere in 1998 there were numerous developments relating to GM crops and food. For example, in February the CGIAR called for a moratorium on the granting of patents on plant germplasm from LDCs. In March, the Australia New Zealand Food Authority rejected mandatory labelling of GM food. In August, a bill seeking to enforce labelling of all GM food was defeated in the New Zealand Parliament, whereas, in the same month, the Japanese Ministry of Agriculture, Forestry and Fisheries issued a draft report supporting the mandatory labelling of GM food. In September, approval given by CTNBio for the release in Brazil of Monsanto's glyphosate-tolerant soybeans was put on hold following a legal challenge by a consumer group.

Farmers in the USA were expected to grow 12.6 million hectares of GM maize in 1999 out of a total of 32 million hectares cultivated for the crop. In two studies in the USA, production of maize and cotton expressing *Bacillus thuringiensis* (Bt) toxins was associated with higher yields and reduced insecticide usage. The National Center for Food and Agriculture (http://www.bio.org/food&ag/ncfap) found that planting of Bt cotton on 17% of the cotton hectarage in 1998 resulted in a reduction of pesticide treatments on 2.1 million hectares such that cotton growers gained $92 million in net income. Maize farmers gained $72 million in net income in 1997, but in 1998 the increased yield did not cover the overall cost of the technology. By examining data from the USDA Agricultural Resource Management Study on the adoption of GM cotton, maize and soybeans in 1996-1998, the USDA Economic Research Service (http://www.econ.org.ag.gov/whatsnew/issues/biotech) noted higher yields and reduced insecticide usage for Bt maize and cotton, but the yield advantages and reductions in pesticide usage were variable.

Following approval by the USDA in 1992, the first commercially grown transgenic crop was planted in the USA in 1994 after clearance was given by the US Food and Drug Administration (FDA). This pioneering crop, Calgene's FlavrSavr® tomato, carried the polygalacturonase gene expressed in an antisense direction. Large-scale commercial plantings of transgenic crops commenced in 1996 with the introduction of Liberty Link canola (oil-seed rape) and BollGard cotton in the USA. By 1998, 22 million hectares of transgenic crops were being grown in the USA alone. Although it is difficult to predict commercial viability, the main transgenic crop releases have been of maize, potatoes, tomatoes, soybeans, cotton, oil-seed rape, tobacco, melon, sugar beet, squash, rice and wheat. Other transgenic crops released included *Agrostis*, *Populus*, *Eucalyptus*, alfalfa, cucumber, grapes, strawberries, lettuce, walnut, sunflower, apple, barley, types of *Brassica oleracea*, and groundnut. According to C James in the *Global Review of Commercialized Transgenic Crops: 1999*, produced by the respected International Service for the Acquisition of Agri-Biotech Applications, there was an increase of 44% (equivalent to 12.1 million hectares) in the global area of transgenic crops between 1998 and 1999. In 1999, it was estimated that 39.9 million hectares were planted with transgenic crops. Seven transgenic crops were grown commercially in 11 countries (USA, Argentina, China, Australia, South Africa, Mexico, Spain, France, Portugal, Rumania, and Ukraine), three of which, Portugal, Rumania and Ukraine, grew transgenic crops for the first time. The four major transgenic crops were soybean (54%), maize (28%), cotton (9%) and oil-seed rape (canola, 9%).

Rapid sophistication is taking place in the objectives or targets of the transgenesis processes. The first generation of crops is predominantly aimed at crop protection by resisting competititon from weeds by the introduction of herbicide-tolerant genes, usually deploying a single gene trait, or by resisting the depredations of pests and diseases. This strategy is still under refinement, extending the range of crops and genes, and addressing a wide range of pests and diseases. Stacking and mixing of genes is being introduced to overcome the possible build-up of resistance in the pest and pathogen populations.

Second-generation transgenic crops are aimed at improving directly yield efficiency and quality. Over the next 5 to 10 years, large-scale crop introductions will probably focus on (i) modified carbohydrate quality for industrial feed stocks (binders, fillers, stabilisers and thickeners) and improved food processing, (ii) vegetable oil content and quality, (iii) amino acid and protein content and quality, (iv) harvestable fibres with low lignin content, and coloured cellulosic fibres, (v) substrates for the bio-plastics industries, (vi) toler-

ance to biotic and abiotic stresses, (vii) better water- and nutrient-use efficiency, (viii) enhanced photosynthetic efficiency, (ix) reduction in the production of anti-nutritional and allergenic factors, (x) modified colours and shapes of fruit, vegetables and flower crops, (xi) easier harvested crops by phenotypic modifications (*e.g.* synchronised maturation, improved abscission) and with improved shelf/storage life, and (xii) hybrid crop production.

Third-generation crops which may be grown on a large scale in the longer term tend to focus on phytoremediation of contaminated land and water, and on the production of nutraceuticals and pharmaceuticals. Environmental monitoring and the slow, methodical approach in carrying out dietary and clinical trials will of necessity delay the introduction of these fascinating crops and platform technologies currently under investigation. The recent interlinking between Biosource Technologies Inc., SCRI and Mylnefield Research Services Ltd represents a major step in the advancement of plant biotechnology to revolutionise the treatment of mammalian diseases. The second- and third-generation crops will have genes that are targeted at specific integration sites in the chromosomes; some will employ organelle transformation, virus vectors, switch technology and eventually also gene-use restriction technology (*e.g.* 'Terminator') which can not only protect the intellectual property by effectively preventing further propagation but stop inadvertent spread of GM or other crops. The problems of gene silencing, location in the genome, resistance breakdown, genetic instability and unexpected pleitropic effects will continue to be addressed by screening in conventional trials, and by technological advances. The destabilising and often misconceived arguments about GM crops and GM food are already influencing investment strategies and may deprive the UK of proper participation in accessing and reaping the benefits of all branches of biotechnology.

At a time when plant sciences in the UK are in marked decline, subject to a few prominent exceptions in the UK public-sector research institutes, such as SCRI, and some universities, the US National Academy of Sciences recommended that urgent attention is given to the fundamental aspects of plant biology, especially molecular, cellular and whole-plant processes, ecology, and interactions between plants and other organisms. Plant science should also incorporate a global perspective. Advances in molecular genetics, mathematics and environmental sciences have enabled the plant sciences internationally to be at their most intellectually and industrially buoyant ever. Nonetheless, a combination of (i) prolonged underinvestment, (ii) unprecedented levels of bureaucratic 'shaping' of research programmes, (iii) under-performance of undergraduate and postgraduate teaching, (iv) poor career prospects affecting recruitment and retention of outstanding scientists, (v) outdated public-sector attitudes that cause a pronounced lack of appreciation, or actual impedance, of technology-transfer initiatives that could introduce new resources and technologies, and (vi) a hostile funding and social environment for plant biotechnology, have all led to the current unsatisfactory position in the UK. Botany is an endangered scientific species. This also coincides with the sad position of most sectors of UK agriculture and horticulture which desperately need to generate profitability by (i) accessing those areas of science, engineering and technology that enable them to be internationally competitive (*e.g.* improved cultivars, greater automation, ownership of intellectual property), (ii) proper customer focus, and (iii) linkage with industries that add value to primary produce. Valiant attempts by the Levy Boards (*e.g.* British Potato Council, Home-Grown Cereals Authority and Horticultural Development Council) are diminished by the economic climate, but their rôles are central to the rejuvenation of their respective sectors of industry, especially if they are not destabilised by continual reviews and are permitted to have an adequate period to participate in generating protectable intellectual property. Fortunately, some areas of agriculture and horticulture attending to niche markets or meeting customer needs are still profitable, and many industries upstream and downstream of agriculture are particularly healthy. Biotechnologies relating to diet and health, forestry, the environment and platform technologies are especially promising.

A dispassionate review would be timely of the rôle of all those various bodies responsible for distributing public-sector resources in UK science. This would include (i) the mechanisms and impacts of their funding on shaping the national programmes; (ii) the scientific outputs and economic impacts of the work they have funded; (iii) estimates of the value-for-money of that science; (iv) the mechanisms, costs and effects of the reviews they have carried out on the structures of the various scientific institutions they fund; (v) the full economic costs of the bureaucratic systems employed; (vi) the social impact of the work funded; (vii) their interactions with international col-

laborators and competitors especially in the EU and USA; (viii) possible, much simplified and cheaper mechanisms to disburse resources; (ix) future mechanisms to highlight priority areas of work and synthesise truly multidisciplinary teams; (x) future mechanisms to connect directly with the public and politicians; (xi) use of internationally robust peer-evaluation systems to review and assist in setting demanding educational and research targets, and (xii) future mechanisms to assist in changing a risk-averse culture.

Until recent times, there were procedural barriers placed by central, interventionist planners and decision-makers to allow public-sector science of all disciplines to flourish in, or interact productively with, the free market (which includes industry, commerce, charities and philanthropists). The August 1999 report by J Baker to the Minister for Science and the Financial Secretary to the Treasury *Creating Knowledge Creating Wealth. Realising the Economic Potential of Public Sector Research Establishments* is a landmark document, illustrating and justifying the pioneering rôle of MRS Ltd and the few other technology-transfer arms established in the late 1980s and early 1990s. Government and senior civil servants and policy advisors are in a difficult position. Are there too many weak or disillusioned, inflexible and/or under-supported scientists and related administrators that remain employed in inappropriate types of activity merely because of the existence of a cushion provided by taxpayers? Undeniably it is an uncomfortable cushion because of the exigency of public accountability, and low reward, but it is sufficiently stable to sustain an educational, research and bureaucratic infrastructure irrespective of diluting the effectiveness of the overall spend. In addressing any fundamental changes, with all the frictional costs involved, it would be false to adhere to the linear model of the economy being dependent on developments arising from applied science which in turn is derived from basic science. Advances in basic science depend as much on advances in technology as *vice versa*, and basic science can reach the marketplace quickly. Indeed, T Kealy pointed out in 1996 that economic growth is technological development. To contribute effectively to social and economic development, science needs an interactive environment of intellectual and procedural freedom, simply because the activity of science is unpredictable, unending, exciting and visionary. Moreover, policy advice, let alone fundamental and strategic science, does not require mental corsets.

In last year's *Report of the Director*, I stated that SCRI has steadfastly sustained a phenomenally productive, pleasant, and forward-looking research environment in the beautiful setting of the Tay valley. Despite the continuing harsh financial retrenchment in the public sector, and enormous stresses on staff, I am pleased to state unequivocally that the Institute, MRS Ltd and BioSS still thrive, producing high-impact scientific research and development with unrivalled value-for-money and productivity, and meeting end-user needs. We play a full rôle in UK and international science, launching major scientific initiatives, and we participate extensively in higher educational activities. I thank SERAD and all our sponsors, and congratulate and offer my gratitude to my colleagues for their loyalty, forbearance and outstanding efforts, not least for their support during a period when I was diagnosed with and treated for lymphatic cancer.

Gene flow in the environment – genetic pollution?

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Biological invasions have had profound effects on human society from the earliest times. The spread of the black death in the Middle Ages, the devastations of potato blight, the effects on indigenous species by grey squirrels, dutch elm disease and flatworms have all been seen as detrimental to man or the environment. Others are seen as bringing benefits: most of our crops evolved elsewhere in the world and many culinary and medicinal herbs were brought to Britain by the Romans. Perhaps the greatest invasion is the import of vast numbers of exotic plants to gardens and greenhouses. Ecological invasions are an intrinsic part of ecology and evolution and we only consider them bad if they impoverish our health, livelihood or living conditions.

Recent events brought awareness of a need to be knowledgeable and vigilant about where food comes from, how it is grown and what it might contain. During the 1990s, agriculture and the food chain returned to be daily topics of conversation. In 1998, the concern was for genetically modified crops, and the term 'genetic pollution' came into common usage. There is no place in science for such emotive terms but publicly funded science must answer, as far as it can, the public's fears with facts and sound interpretation.

The questions? Biological science is regularly in the spotlight. Is habitat being destroyed by land management practices? Will GM crops escape from fields, affect plants and animals, injure us? Will crops in general, and GM ones in particular, reduce even more the biological diversity of arable farmland? Will they contaminate other crops, cause more pesticide to be used, rather than less as some companies claim?

An increasing number of people have a stake in the debate - pressure groups, farmers, farm advisers, consumers, agrochemical companies and government. Opinions are too often polarised. In this confrontational atmosphere, the need is for clear, independent fact, answers and comment. A part of the debate is ethical, but independent research is essential on questions that science can legitimately address. This is not to say that scientists should not be ethical, but that the methods of science can only be applied to answer certain types of question through constructing hypotheses and testing them by observation, experiment and modelling. It cannot say, for instance, that grey squirrels or GM herbicide-tolerant crops are 'good' or 'bad'. The answer depends on your standpoint. Science can, however, define what effect such organisms are having or might have on wildlife, habitat and health, and advise government and the public on the risks and hazards.

Invasions are complex biological processes. How do we study them? First there is the movement of plants or seeds, and the transfer of genes by pollen. The organisms have to find and occupy space and grow in their new environment, while the pollen has to find

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receptive stigmas and create a new organism. Incoming plants have to interact for space and resource with the plants that are already there, and accumulate enough mass to reproduce and maintain a stronghold. The local environment acts on them and sorts out the less able. Exchange of genes creates variants that are continually re-sorted. The nature of the invaded population changes from what it was at the beginning.

The difficulty lies in the range of interacting scales. Growth or death, and exchange of genes, occurs through direct interactions between individuals. The individuals occur in patches or fragments and the fragments are scattered over the landscape. Single fragments evolve their own characteristics that fit them to the local environment. The patches are not independent, but potentially connected through the migration of seed or transfer of pollen. The individuals in distant patches can therefore influence the interactions within any other patch. Clearly, the degree of connectivity will depend on whether the pollen or seed moves by wind or is carried by insects or other animals, and how sexually compatible are the plants in the different patches. This system evolves therefore at scales of both the patch and landscape. This is what makes the study of gene flow and invasions so difficult and so challenging. Very advanced techniques are being developed and used to examine these exchanges.

New methodologies Movement of seed or genes into a fragmented ecosystem usually begins slowly and proceeds gradually. Only when things are well on their way are they generally noticed. The crux is detecting and interpreting rare events and small numbers. Methods and techniques are needed to do three things: to show that an organism or gene has moved from one place to another; to measure its effect on the existing organisms at that place; and (because these two can only be done on a small number of the organisms) to build the greater picture from all the available data. The sciences of plant ecology, genetics, zoology, pathology, mathematics and statistics are all put to use in this work. Research not only has to break new ground in each subject but to find new ways of seaming them together.

Genetic exchange and detection Advances in knowledge of the genetic code have led to robust techniques for genetically typing individuals and for detecting the movement of genetic material from one plant to another. Some of the techniques now in use at the Institute[1,2] are shown in Figure 1. Each is appropriate for a specific purpose. To confirm geneflow, it is essential to observe a transfer of genetic material from one individual or group to the progeny of another. Potential receptors and donors need to be genetically typed to search for distinguishing features in their DNA. Seed is collected from the potential receptor (mother) plants, germinated and the seedlings also genetically typed. The DNA of the mother plant and seedlings are then compared. Gene flow has occurred if a particular DNA sequence is found in a seedling but not in the mother plant. Further, if the number of potential donors is reasonably small, then some of the techniques can trace the plant from which the pollen came. As Figure 1 shows, the approach is being used to detect and quantify gene flow in plants ranging from feral brassicas to tropical trees.

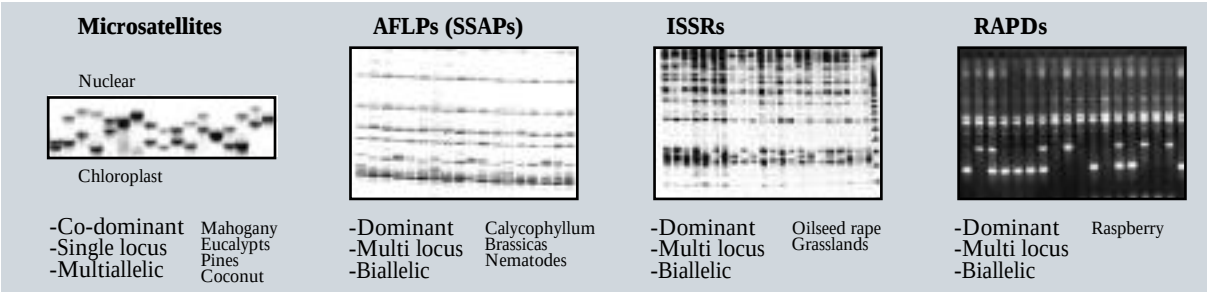

Figure 1 Banding patterns from some genetic technologies, with which it is now possible to assess gene flow and the spatial organisation of genetic variation. The techniques are used at SCRI with the species indicated.

The new genetic enabling technologies allow us to do things that were impossible a few years ago. To answer most questions, further knowledge is needed of how seed or pollen are moved by wind or insects, and whether plants are sexually compatible. This brings in new scales of operation that are influenced by the local weather and the flight paths and foraging behaviour of pollinators. Molecular science can contribute to knowledge at these scales by helping to define the distance over which pollen can move from one plant to another.

Science in this area has recently made major contributions to the topical and contentious issue of gene transfer from GM crops to wild plants. It has been suggested that GM characters will be transferred to weeds giving them resistance to weedkillers and insect pests. The flow of genes from some crops to some wild relatives is possible and will happen if GM crops are deployed on a large scale. Indeed, crops and weeds have exchanged genes since the beginnings of agriculture, so there is nothing new in the process itself. So far, there are few indications in the UK that crops are crossing with other species to increase the latter's ability as a weed. Those crops and wild relatives that are most likely to exchange genes in the UK are known in principle[3]. For instance, the potato has no wild relatives in the UK that are sexually compatible with it, whereas oilseed rape can potentially cross with wild turnip and wild radish. However, gene transfer is much more likely from crops to feral descendants of crops growing as weeds within fields, and could cause problems for the farmer.

Economic issues aside, any transfer from crops to wild relatives might impoverish their diversity, an event unwelcome in itself but which could weigh against crop improvement in the long term if the wild relatives contain potentially useful genes. Again, sexual compatibility is crucial in assessment. The risk in Tayside of cultivated strawberry crossing with wild strawberry is negligible because they are genetically incompatible, while that of cultivated raspberry crossing with wild raspberry is moderate because they can mate freely. Detailed studies at SCRI on wild and cultivated raspberry are given later as a case history.

Sorting, extinction, amplification Seeds or vegetative parts of invasive individuals face not only severe physical and chemical environments but also the competitive influences of other organisms. Seed is attacked by fungi, bugs and birds. If it lives to germinate, it faces an aggressive set of species. An invading organism enters an ecological assemblage that primarily determines its rate of establishment, extinction or amplification. Many factors mitigate against the ingress of an alien organism, and many invasion events and many cycles of reproduction are likely to be required before it gains a secure place.

Invasions and population structure In an established arable field or wayside patch of ground, perhaps 30 weed species cohabit in the soil as dormant seed. Among them are likely to be several of the typical, economically damaging, arable weeds: fat hen, cleavers, blackgrass, wild oat. Each is at least as capable of exploiting time windows in the weather and husbandry as any invading type such as feral oilseed rape or feral beet. The existing seedbank species occur in some hierarchy of abundance, and are distributed over space in characteristic ways. Typical abundance rankings show a wide range from species that are very dominant (10,000 to 100,000 in a square metre down to plough depth) to those that occur relatively scarcely (<1000).Which individuals are dominant depends on the soil, the rotation, the use of herbicide, and other factors. The Institute's unrivalled database on arable buried seed (seedbanks) is continually probed to answer questions on arable diversity[4]. Spatial maps of the seedbank in a field (Fig. 2a) have interesting features, but interpreting 30 or more of these for individual species presents a major problem. Statistical procedures are therefore used to condense the masses of data that accumulate from measuring communities. For example, Principal Co-ordinate Analysis gives a quantitative and also visual summary of the diversity among samples or experimental treatments. The analysis is applied in Figure 2b to compare the assemblage of species that developed in two rotational treatments at three sites[5]. Each symbol represents the arable plant community at a sample point in a field. (The farther apart two symbols are, the greater the difference in the species detected at the two sample points.) The seedbank was small and similar at the start of the experiment (not shown). After 6 years, during which herbicide input was reduced and spring-sown crops were introduced to the winter cereal rotation, a marked divergence of the communities occurred, shown by the spread of symbols and separation of sites. The weed communities were very different at the end, potentially providing contrasting opportunities for an invading species. Land management and location are therefore central to the progress of invasions.

The feral weeds of crops such as oilseed rape, beet and potato, and certain wild relatives, all now inhabit

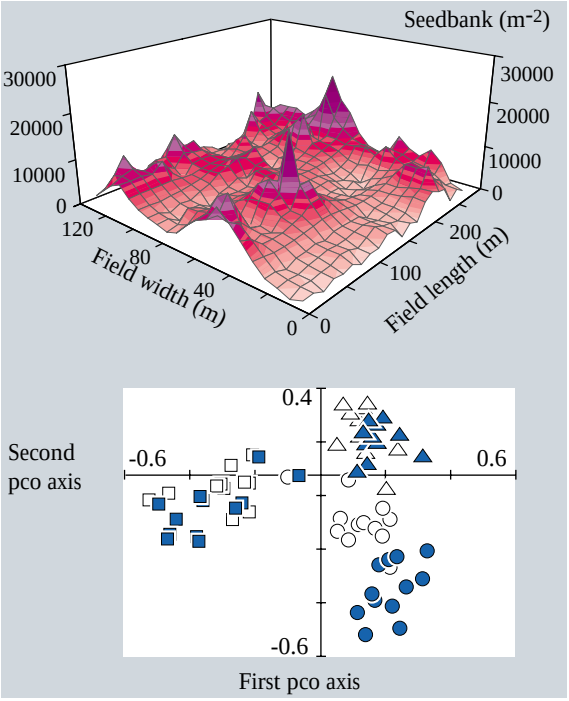

Figure 2 Examples of (A) spatial pattern in a weed community and (B) Principal Co-ordinate Analysis, which condenses information on many species: symbols represent data for experimental plots at three sites (triangles, circles and squares) and two rotations per site (open and closed symbols). The distance apart of any two symbols is a measure of the difference in the species present.

seedbanks of arable and waste land. The significance of the feral plants is that they will emerge with a crop of the same species, are largely indistinguishable from the crop, are difficult to control in that crop, are generally sexually compatible with it, and are close enough to cross-pollinate with it in quantity.

Linking fragments in the landscape The Institute now has an extensive network of sites throughout the UK and overseas where the composition and dynamics of plant communities are studied. More intensive research to link both fragment and landscape scales is concentrated in the Tayside region using 'model' plant systems: the feral brassicas[6], descended mainly from swede and turnip oilseed crops and the wild raspberry[7]. Additionally, and as part of co-ordinated research programmes, the Institute carries out field studies on species-rich grassland, Scot's Pine[8] and a range of tropical trees. Populations are mapped spatially and their origins and persistence inferred. In selected fragments, the genetic markers are used to discover which cultivars they descend from and whether new genes are entering the fragment. This long-term work of plant demography provides a natural laboratory, a sound foundation and a context in which to answer a range of questions on policy and management (examples are given later). Because all these populations are examined with minimal disruption to them, further science is needed to understand how they function.

Detection and quantification of pattern It is imperative to be able to detect small changes in populations so that the impacts of migrating individuals or incoming genes can be judged. It is not usually enough to detect that a particular event has occurred. Migrations and other invasive events have spatial and temporal dimensions: they affect the patterns in existing populations and not just the numbers. For this reason, techniques of spatial statistics are being developed which allow an existing pattern and its change over time to be quantified[9]. The technique is demonstrated by means of four artificial patterns, which were simulated by a computer programme (Fig. 3). The blue and pink squares could, for instance, represent individuals of a different genetic type, and the intensity of colour, their numbers. In the top left pattern, the colours are independent of each other. In the top right, they are arranged along a gradient with more

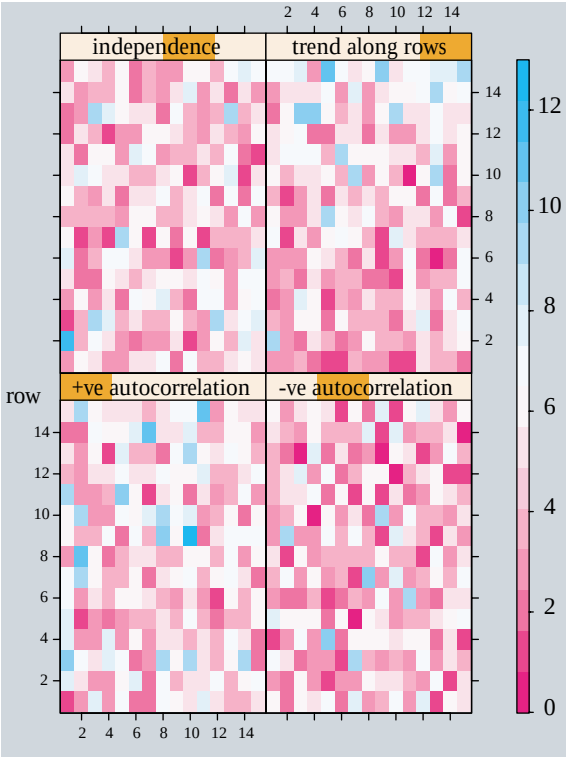

Figure 3 Four computer-simulated patterns to show different types of spatial arrangement between pink and blue 'organisms', the abundance of which is indicated by intensity of colour[9]. Text gives explanation.

pink at the top and more blue at the bottom. The two graphs at the bottom show different forms of distribution where the colours and intensities are not independent: deep pink squares are commonly adjacent on the right, rarely adjacent on the left. Similar patterns occur in nature and tell us something of the underlying processes. It is not easy, however, to establish by eye that the four patterns are different, and in particular, independence (top left) looks very similar to positive autocorrelation (bottom left). There is a clear need, therefore, to demonstrate and quantify spatial pattern and its change over time using such advanced statistics. These methods are now being applied to vegetation and will find many uses in population dynamics and gene flow.

Imposing pattern experimentally A further, complementary step is to study the way artificial patterns affect interactions between plants. Much previous work in cropped land has examined plants in fairly uniform arrangements. Experiments on crops typically vary the density (number of stems per square metre) but keep plants regularly spaced. This is not how weeds and other wild populations become arranged. Rather, invading individuals find themselves within complex existing structures of different size, density and shape. Trial designs that simulate more variable states are needed for this purpose.

As examples, three planting patterns are shown in Figure 4. Each represents a feral or weedy patch having the same number of plants but a different degree of aggregation. From left to right, the pattern gets more aggregated, with more open space, more crowding, and more internal 'edge'. The basic pattern is a Sierpinski triangle, whose various forms allow precise computation of inter-plant distance and plant neighbourhood. Comparing a number of designs like this enables us to look at both competition and gene exchange over a wide range of plant neighbourhood in a (relatively) very small space. The design illustrated was used in 1998 to examine the exchanges that might occur if a new cultivar of oilseed rape (pink squares) invaded an existing patch (blue squares). The more aggregated patterns caused greater size inequality among the individuals. Also, molecular detection techniques showed that up to 60% of the pink progeny contained genetic information from the blue, suggesting that the type in low number was diluted by the dominant type. In this experiment, the types were two different conventional varieties, but the technique can be adapted to investigate the influx of new types of crop. This is fairly fundamental science that will help our understanding of applied work on the spread of genes among feral crop plants. Research using these imposed patterns provides a link to understanding the real happenings in nature.

Mathematics for scaling-up and management

When a gene is modified by conventional or GM technology, it causes certain changes in the properties of the plant. However, many questions about that gene are asked of its effects at the much larger scales of the plant community or landscape. Will it alter the use of pesticides or the diversity of arable flora and fauna? Is there some state in the regional distribution of vegetation fragments that gives rise to rapid invasion and gene flow, and can land management act to prevent this state occurring? Do things happen at the landscape scale that are simply not evident from research on small scale trials? Very often, the full information required to answer such questions is not available. Pieces of the picture exist but not the whole. Research funding is limited, so where should the effort go?

At SCRI, we are developing advanced mathematical techniques both to link disparate sets of information and to form hypotheses to aid our experiments. The sequence goes like this: certain patterns (in species

Figure 4 An experimental design based on a Sierpinski triangle, used as a planting pattern to study gene flow and competition among two plant types (identified by colours). The number of plants in the patch remains the same but the degree of aggregation increases from left to right.

abundance, spatial distribution, etc.) in a vegetation type have been detected in the field. The origins of these are uncertain so they cannot be managed. If the community is not probed, its functioning will be obscure; if it is disturbed, it might show untypical effects. As an intermediate step to gain further knowledge, a limited number of plants are removed and their physiology and genetic diversity investigated. Next, simulated communities can be set up (as above) in imposed patterns. But despite all this, there is still uncertainty. Mathematical models are therefore used to reconstruct the community from all the knowledge there is about it. The point of modelling is that it predicts something about some feature of the community that can then be tested with minimal interference back in the field. The process of modelling and experiment is cyclical.

In the most advanced models, individuals interact through physiological processes over a spatially and temporally variable resource (simulating nutrients). Change over time shows a succession from plants that seed rapidly (typical invaders) to plants that retain resource. In this way, models show how the interactions between individuals affect the evolution of patches, and how invasions might occur and be checked. Research is moving rapidly towards simulating the effect of introducing new botanical characteristics (including those of GM plants) to the modelled community. Other forms of modelling work at the scale of the landscape to examine how genes might move between fragments or from crops to feral plants and weeds[10, 11].

Questions answered on conservation and ecological risk *Conservation of fragmented populations* The spread of urban land and intensive agriculture and forestry during the past two centuries has caused wild places to decline in area. Many wild plants now exist in fragmented habitats or patches sometimes separated by long distances. Modern approaches to conservation demand that such habitat is at least maintained and where feasible increased. However, the conflict of different interests means that questions are often asked of the importance of a plant or a group of plants. For instance, how much of the diversity of a species will be removed if a certain habitat is destroyed or how much habitat needs to be conserved if the diversity is to be maintained? The techniques described are now being used to answer these practical questions. A local and a more exotic example illustrate the potential.

The wild raspberry (*Rubus idaeus*) grows throughout Europe, including the raspberry-growing areas of Scotland where it is important to the economy. The wild and cultivated raspberry can cross-pollinate and exchange genes. Questions arise about the diversity of the wild populations, whether the cultivated raspberry is eroding the diversity of these populations, and whether the wild populations contain genes and character traits that might be useful in breeding for, say, pest resistance or hardiness to cold? One obvious difference between the two forms is that the wild are generally spiny while the cultivated have smooth stems. A search for smooth stems in wild populations[10] suggested there was little escape from plantations to the wild. However, spininess is not a neutral character, in that spine-free escapes might be more likely to get eaten than spiny ones. Accordingly, a combination of DNA-based[7] and physiological methods were used to explore the diversity of wild populations compared to the widely used commercial clones.

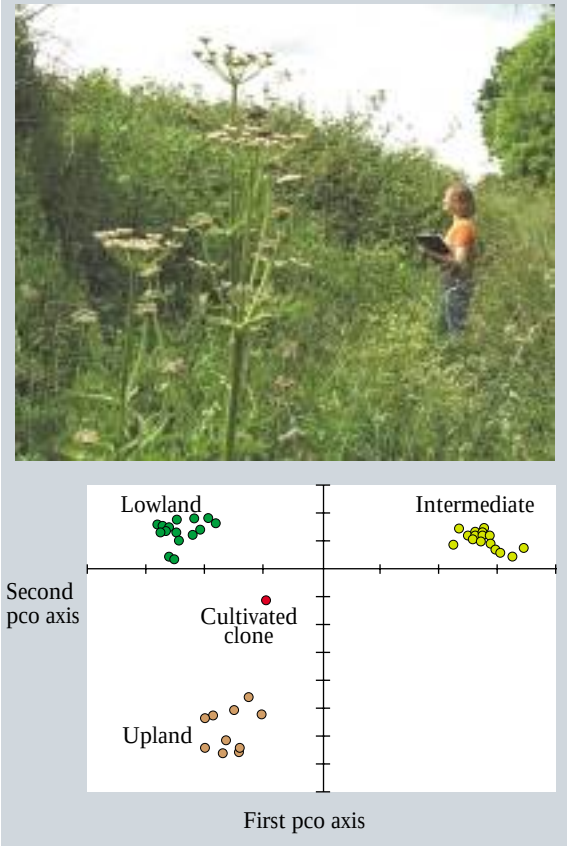

Figure 5 Statistical analysis of genetic marker data (RAPD, Fig.1) of wild raspberry individuals collected from upland, lowland and intermediate sites; distance between symbols (individuals) is a measure of genetic dissimilarity and clumping shows genetic differentiation between sites. None of the populations is similar to the cultivated clone. Photograph shows a lowland population.

The results showed evidence of great diversity in the wild populations compared with the cultivated clones. The study is not complete, but results so far indicate there is not much gene exchange between the wild and cultivated forms. Are there areas of greater diversity? The most diverse forms occurred in the Angus Glens, but even wild populations near the Tay estuary were quite different from the cultivated clones (Fig. 5). If the upland populations were erased, then certainly much of the diversity in form and genotype would be lost. They have an important ecological role, providing cover and food, but we do not yet know whether these wild populations contain genes and characters that would benefit the soft fruit industry. A collection of wild raspberry has now been established at SCRI, for the purposes of study and conservation.

The second example is a dispersed population of the tropical hardwood mahogany, *Sweitania humilis*, where the remaining trees occupy isolated patches in the landscape. Pollination occurs by means of an insect. Questions were asked whether isolated single trees or small groups were genetically cut off from the rest and how their conservation should be managed. They might, for instance, contain important genes and characters but not be able to contribute these to the larger patches, some of which lay several kilometers distant. A DNA marker technique was developed[11] which enabled the genetic material carried in pollen from each individual tree to be identified. If a DNA marker from one tree appeared in the seedlings produced by another tree, then clearly, genes had moved with pollen from one to the other. The results showed there was extensive gene flow between patches and that isolated trees were still contributing to and receiving genes from the larger patches (Fig. 6). Keeping the isolated trees was therefore important for conservation of the species in that area. Much is being learnt from this work and that on wild raspberry, about the kinds of fragmentation pattern that encourage genetic exchange and diversity.

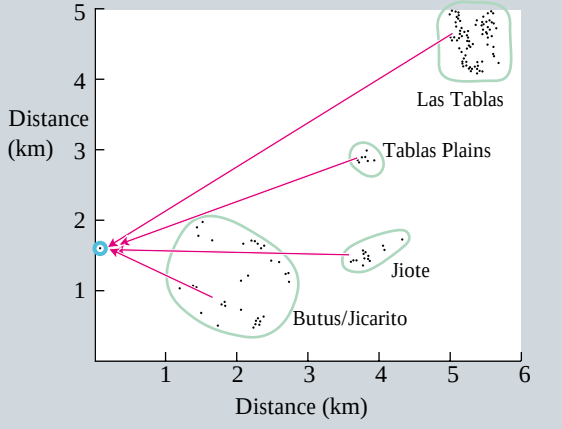

Figure 6 Gene flow to an isolated tree of the mahogany, *Sweitania humilis*, from fragmented patches up to 5 km away (using microsatellites, Fig. 1)[11].

Risk assessment and GM Crops Since 1993, work at SCRI on the assessment of ecological risk has concentrated on oilseed rape. A number of important questions were asked by government concerning the distance travelled by pollen and genes, the persistence of feral populations, the contact between ferals and crops and the general likelihood and extent of gene

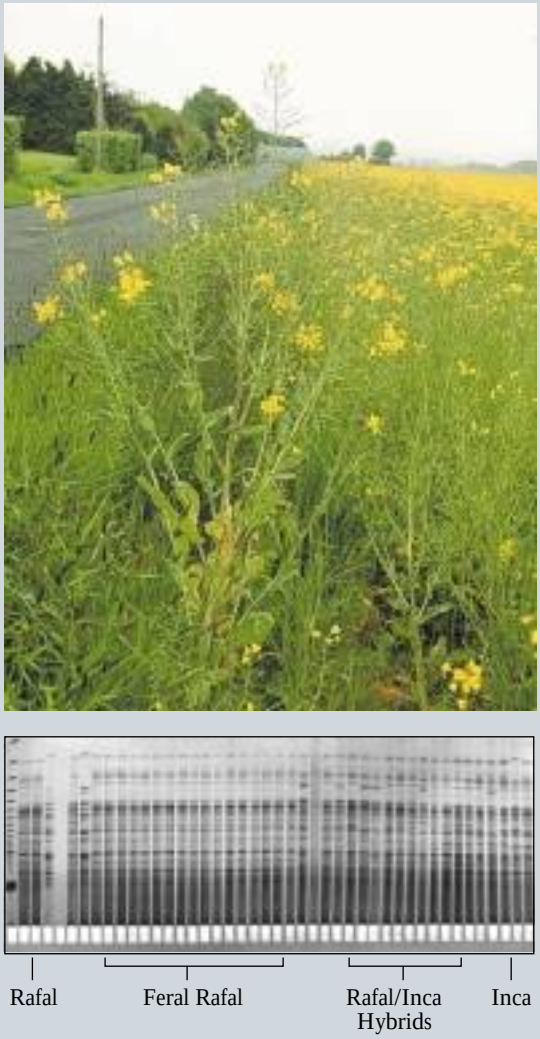

Figure 7 Feral oilseed rape population (large plants left) growing adjacent to a crop field in 1998, and DNA fingerprints (ISSRs, Fig. 1) of two varieties, Rafal (grown 1987) and Inca (grown 1998), and of a selection of the feral progeny in 1998 showing the Rafal fingerprint and Rafal-Inca hybrids.

flow across an agricultural region. One outstanding question was settled in 1998, and that was whether a feral descended from one crop type could hybridise in a subsequent year with a new crop cultivar. It was previously shown that the predominant DNA fingerprint in a persistent feral population was equated with a source crop last sown in autumn 1986. Other genetic types, possibly other varieties and hybrids were present also in the population. However, corroboration of gene flow was obtained by showing that an adjacent oilseed rape crop, flowering in 1998, hybridised with the feral descendants of the original cultivar (Fig. 7).

Moreover, the integration of data and theories through mathematics enabled a finer definition of the problem at the scale of the landscape. The question that modelling addressed was whether earlier measurements from single fields near SCRI could be scaled up to predict gene flow in a landscape of tens or hundreds of kilometres. Different sets of data were combined with a physical model of pollen dispersion to give the first regional scale model of gene flow, which predicted (contrary to much current thinking at the time) that most feral populations could receive genes from several fields and that the regional aggregation pattern of the fields determined how much each population would receive from a hypothetical GM field. Fields and ferals were therefore 'regionally coupled'[12, 13].

This prediction of course, required verification. Accordingly, male sterile bait plants, which do not produce their own pollen and which act as biological pollen detectors, were dispersed around the countryside during the oilseed rape flowering season in 1998. Any seed set on these plants indicates pollination by other plants. This large-scale approach in an agricultural landscape was a radical departure from previous methods. It confirmed that pollen indeed travelled long distances, that bees were important vectors between crops and ferals, and crucially (by taking advantage of the presence of a commercial GM trial in Tayside), that the spatial arrangement of GM and non-GM fields determined the balance of GM genes in the progeny of the bait plants[14, 15]. Because the bait plants do not produce their own pollen, uncertainty remains in the true magnitude of gene exchange and establishment of hybrids at distance from a source field. Research is continuing to examine the relative ability of self and foreign pollen to fertilise flowers and, in effect, to calibrate the data using bait plants.

The progression of research on the ecological effects of oilseed rape [6, 12-16] would not have been possible without the contribution of many disciplines. The main findings can be summarised as:

- pollen moves over at least 4 kilometers;
- the pollen is still alive at these distances and can pollinate flowers;
- it can be carried by both wind and insects;
- many feral populations die out quickly but others persist in fields and around agricultural land for at least 10 years and probably longer;
- feral populations can receive genes from crops of oilseed rape.

The implication is, that by these means, genes from GM crops will likely enter feral oilseed rape populations and persist there for several years. However, the ferals are unlikely to dominate the arable community. For example, feral oilseed rape occurred at all three sites in Figure 2, but did not take particular advantage of the new niches as did many other weed species.

The need to explain complex and uncertain information

The public increasingly want to be informed and reassured about risk and hazard. Scientists must find ways of getting the message over, despite the various grades of certainty in the data. Some conclusions can be stated with absolute certainty and these are the easiest to transmit. The statements on oilseed rape bulleted above are all-or-nothing statements. They are very useful for giving government departments and the informed public the "worst case scenario". Even though quantities are included (4 kilometres, 10 years), none of the statements gives a measure of how much will occur or how likely each event is to happen.

Others conclusions have to be qualified by some expression of uncertainty. Moving to the question of how much pollination or gene flow there was at such and such a distance, there is no certain answer. The result depends on many factors: local weather, the lie of the land, the number and behaviour of insect pollinators, the size of the source field, the size of the sink population, and the arrangement of surrounding fields. The answer to a question of this type is therefore much less straightforward but can be given scientifically by means of graphs and cautiously worded arguments stating statistical probabilities. In order to address this problem, a research area is developing at the Institute specifically on modelling and interpreting uncertain information[17].

Admittedly, this advanced uncertainty-modelling is unlikely to enable the non-scientist to grasp the risks and hazards. Other forms of wording are needed. Few of us can cope with too many categories; perhaps no more than three, such as certain to happen, likely to happen and unlikely to happen. Examples of certain to happen are given above. That genes will move from GM oilseed rape crops to some relatives is likely to happen. Take again the scare of 'superweeds': the notion that herbicide tolerance will spread from GM crops to arable weed species and create new weeds that are very difficult to control. The weight of evidence is that (although such hybrids are likely to happen), GM superweeds are unlikely to arise in the UK for the following reasons: (a) the wild relatives in question are not particularly aggressive or competitive as weeds, (b) having herbicide tolerance will not alter this aggressiveness unless the particular herbicide is used, (c) other herbicides or weed control practices could be used to control them, (d) other highly aggressive weeds are more likely to dominate in any case.

The implications for agricultural systems The movement of seed and pollen will have to be measured and managed much more in the future than it has been. This will be so whether GM crops are grown commercially or not. Crop products are becoming more specialised and there is an increasing general need for purity. Large sections of the public are insisting that agriculture protects and enhances the habitat and the wider environment.

In principle, and as shown above, detection of gene flow from a GM or other specialised crop into a conventional crop is straightforward. The novel gene can be detected in seeds, or seedlings grown from them, by PCR-based methods. However, when GM pollen from an extraneous source is competing with a much larger concentration of pollen from within the conventional crop itself, the proportion of seeds acquiring the novel gene may be very small. Thus, although PCR-based methods are extremely sensitive, there will be a lower limit below which gene flow will not be detected in practice.

The detectability of gene flow has acquired considerable political importance in the context of food labelling, because consumers are demanding that food derived from GM crops be labelled as such, so that they can choose whether to buy it or not. In a crop such as sweet corn, gene flow from a nearby GM maize crop could result in a few kernels on an otherwise non-GM cob being GM. A scientific question is: at what level is such "contamination" detectable? But there is also a political question: what level is acceptable? The Soil Association, representing organic farmers, wishes there to be a zero tolerance of GM gene flow into organic crops, but this is probably impracticable. A draft EC Regulation proposes a 1% tolerance level for the adventitious presence of GM-derived material in any food ingredient from a non-GM source, for labelling purposes.

In a crop such as sugar beet, in which the yield is vegetative, the product will be unaffected by gene flow into the crop itself, but gene flow in the previous generation into the seed crop may be relevant. Moreover, with a product such as refined sugar, which contains neither nucleic acid nor protein from the source plant, there is no analytical way in which to determine whether it is derived from a GM crop, a non-GM crop, or whether gene flow has occurred in the crop. The consumer's "right to know" may be satisfied only by an elaborate and costly system of traceability.

An interesting sidelight on gene flow occurs when the companies that have developed GM crops try to enforce their prohibition on farmers saving seed from them. Companies will have to counter the defence that the seed were saved from a conventional crop, which had acquired the novel gene by natural gene flow.

Managing geneflow Such issues highlight the fact that gene flow will increasingly be a factor that has to be monitored and managed in daily agricultural practice. Isolation distances that cause detectable or acceptable gene exchange might need to be revised as our understanding of regional processes increases. Much greater co-ordination of planting between farms is likely, as happens already in certain parts of the UK[18], both to reduce genetic exchanges and to manipulate seasonal habitat. New agronomy should result in greater diversity within and around the crop if cultivar mixtures are more widely grown and refuges more widely introduced for natural predators of insect pests. Whatever new technology comes in (GM or otherwise), its effects on the existing biodiversity have to be assessed, and land husbandry altered accordingly.

In 1999, a new wave of research began at SCRI to tackle these issues. One thrust is through mathematical modelling which allows the prediction of the risk of introducing a specified genetic type without necessarily deploying GM crops. Models to understand how to suppress herbicide-tolerant feral crop plants

and to prevent antifeedant genes harming non-target organisms are in this category. At some stage in the procedure of risk assessment, models and small trials are not enough. Given initial appraisal of safety, farm-scale trials are required to confirm or examine ecological processes or possible emergent properties. The farm-scale evaluation of the effect of GM herbicide-tolerant crops on arable diversity is of this type. These government-funded trials, carried out by a consortium comprising ITE, IACR and SCRI, will lead to new definitions of arable ecosystems in the UK. They are, moreover, an example of 'open' research in a highly contentious area.

Fitting the cultivar to the system. There is ample scope also for the plant breeding industry to concentrate more on properties that discourage feral descendants from persisting and spreading. For instance, the proportion of secondary dormancy (which encourages overwintering of feral populations) differs greatly between oilseed rape cultivars. This secondary dormancy occurs mainly when the imbibing seed is exposed to low temperature or water stress, so is less likely to occur when seed is sown as a crop than when it falls from feral plants. For crops such as oilseed rape, potato and sugar beet that leave ferals, breeding should consider the ecological as well as the agronomic properties of its new cultivars. The deployment of 'terminator' technology and engineered sterility, assuming they can be made to work reliably, is contentious and requires further ecological appraisal. Research is just beginning in some highly relevant topics, while greater effort is appropriate in some other areas.

Future questions – new science The concerns over GM crops have brought to attention the need for science to understand agricultural systems, not only the crop varieties or the specific genetic modification. Land has more than one purpose and science must help management optimise several functions rather than maximise one. Yet behind the intensity of the GM debate, two factors are repeatedly implicated. The first concerns the impartiality of the science and the scientists that are carrying out ecological risk assessment. While science must work with industry, it has to retain a degree of independence through public funding. Otherwise, people will not believe its findings on these issues. The second is that the information required to answer the most pressing questions of recent times is seldom immediately available. Too much 'firefighting' research is put in place in an attempt to provide the answers. The solution is a much sounder infrastructure for studying the flow of individuals, species and genetic information across the countryside.

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Cereal variety mixtures reduce inputs and improve yield and quality - why isn't everybody growing them?

A.C. Newton & J.S. Swanston

Cereal variety mixtures, i.e. several varieties of the same species such as barley, sown mixed together, offer many potential benefits to the grower, namely:

- yield increases of between 5 and 15%
- reduced pesticide inputs
- improved grain quality
- stability of yield and quality[1]

Unfortunately, agricultural and industrial end users have concentrated on certain perceived disadvantages, so mixtures have been deployed infrequently in UK agriculture. Recent research, however, offers further evidence of the advantages of mixtures, and new methods to overcome the perceived problems, so it may shortly become opportune for the grain trade to re-consider its position.

How do variety mixtures work?

Disease control One of the main ways cultivar mixtures reduce inputs is by limiting spread of disease, thereby reducing or eliminating the need for pesticide applications. In the case of a pathogen such as mildew, only specific races with matching virulence (lack of an avirulence gene product which can be recognised by the host) can infect a variety with a specific resistance gene. If that race does not have matching virulence to another variety in the mixture with a different resistance gene, then it will induce resistance in that variety. It cannot therefore grow and produce spores for further infection on the resistant variety, and the resistance induced will even reduce the amount of infection by other normally virulent races on that variety. The epidemic is therefore slowed by two spatial effects in addition to the induced resistance; there are fewer susceptible hosts and the resistant host variety provides a barrier to reduce successful transmission of spores to the next susceptible host variety.

Variety mixtures are also effective against pathogens such as *Septoria tritici* and *Stagonospora nodorum*, which do not have the highly specific gene-for-gene interactions common in biotrophs such as mildew. Mechanisms to explain their effects still depend on the disadvantageous effect on a pathogen of changing from one host variety to another resulting in reduced infection overall.

Yield Reduced disease would be expected to result in a corresponding yield increase, this being the primary objective of the use of mixtures, but, in practice, the use of variety mixtures often gives more yield increase that would be expected from the level of disease control. Clearly, the yield response is not simply due to a reduction in the loss of grain filling through assimilate diversion to a pathogen or damage to photosynthetic capacity. It is also due to competition and yield compensation effects within and between the components of the mixture. Whilst a mixture may be sown as, for example, three varieties in equal proportions, the harvested grain proportions may be considerably distorted if a particular component is highly competitive. If that component is intrinsically low yielding, this could even result in reduced yield compared with the mean of the equally weighted components as sown. More often, however, the yield is greater, not only through the same effect of a competitive variety, but also through better resource exploitation overall. A single cultivar may not exploit all the available root or aerial environment for nutrient and light capture at any one time. Within the heterogeneous components of a mixture there is likely to be a component ready to exploit the available resources much more of the time, to the overall yield benefit of the mixture.

The effect of increased yield and disease reduction might be expected to show some relationship to the degree of heterogeneity in the mixture (Fig. 1). Heterogeneity is most easily manipulated by changing the number of component varieties and indeed there are

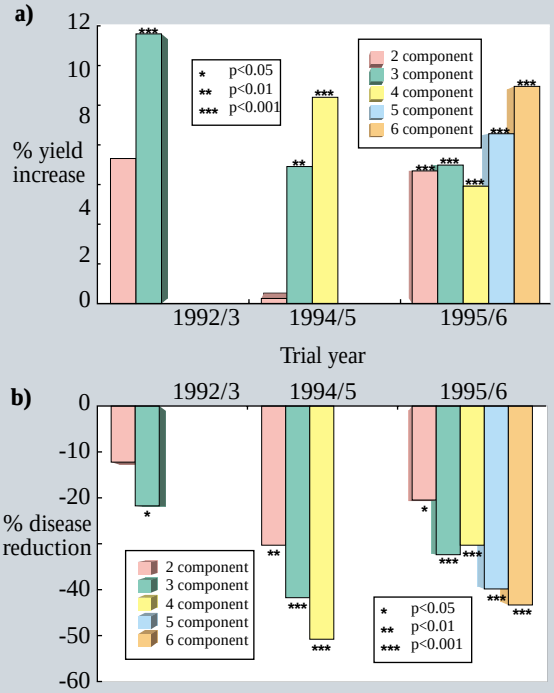

Figure 1 Reduced *Rhynchosporium* infection and increased yield corresponding with number of component varieties in winter barley mixtures. a) Increase in yield in relation to mixture component number. b) Disease reduction in relation to mixture component number.

significant correlations between increased component number and both disease reduction and increased yield, indicating that the higher the number of components the better[2]. However, a large number of varieties is impractical on farm and the number of agronomically compatible varieties available is limited. Whilst the performance of two component mixtures is somewhat variable, three component mixtures are both more reliable, practical, and achieve a high proportion of the disease control seen in much more complex mixtures.

Quality Barley cultivar mixtures are currently grown in Scotland but only as a 6-row high yielding feed quality cultivar mixed with a 2-row to increase the specific weight, i.e. to increase quality. They also tend to reduce lodging. In general, it has been assumed that quality factors such as those required for malting, will be reduced in a mixture compared with the mean of the component varieties. Nevertheless, until our recent studies, there has been very little published evidence to verify or contradict this assumption and several countries, such as Denmark and Poland, have found mixtures can be quite satisfactory for brewing. Our research also indicates that they can be very advantageous for use in malting.

Objections to mixtures

Increased heterogeneity in the malt Maltsters have been reluctant to use barley mixtures for three reasons: increased heterogeneity, verification problems, and customer preference. The first arises as maltsters 'fine tune' their systems to gain optimum performance from a specific cultivar. Therefore, the imposed regime will not be best suited to genotypes which differ in their rate of malting. Too rapid modification, or breakdown, of the endosperm structure will lead to the loss of fermentable material to the growing embryo (malting loss). Slow modification will not degrade sufficient cell wall material and protein to permit ready access of enzymes to all the starch. In addition, cell wall residues can cause viscosity problems and poor filtration. Mixtures are perceived as giving an uneven or heterogeneous malt due to differences between components in malting behaviour.

Verification and customer preference A system which permitted mixtures would require to be effectively controlled. The components and their proportions should be quantifiable and the mixture should be demonstrably what it is claimed to be. Distinguishing between grain supplies of different cultivars may be very difficult using morphological characters. Electrophoresis of storage proteins (hordeins) enables grouping of cultivars rather than individual cultivar identification and is useless for malt samples as storage proteins are substantially degraded. Both domestic and export markets favour monocultures and sales maltsters are frequently required to give assurances to customers. Mixtures are, therefore, not easy to sell at present, but there is considerable evidence that questions the validity of these objections.

Validity of objections For most malting parameters, mixtures give equivalent results to the mean of their components, so any components of inferior quality significantly increase heterogeneity and adversely affect other aspects of malting performance. However, experience, especially in Eastern European countries such as Poland and East Germany, has suggested that mixtures of malting quality cultivars can give acceptable performance in both maltings and brewhouse.

Varietal purity does not guarantee homogeneous grain samples. Even within a field, there may be differences in drainage and soil type, while grains may differ depending on position on the ear or location on main or side tillers. Maltsters do not work with homogeneity, but within an acceptable range of heterogeneity. The range of heterogeneity in well designed mixtures

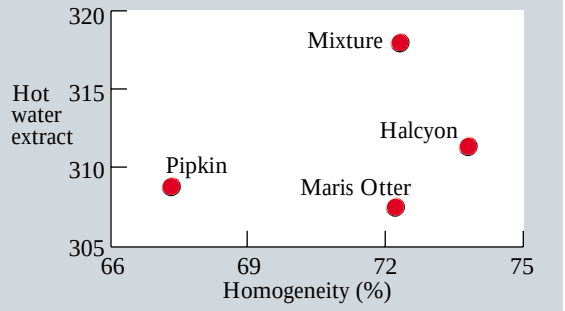

Figure 2 Improved hot water extract and homogeneity of cell wall modification in a mixture of winter barley varieties Pipkin, Maris Otter, and Halcyon.

is likely to be less than the heterogeneity amongst grain from the same variety grown in different places.

Work at SCRI, with a mixture of three cultivars derived from similar pedigrees, showed no significant increase in heterogeneity compared to any of the monocultures (Fig. 2). This was determined by a laboratory test for the evenness of modification of cell walls (Fig. 3). In addition a slight, but significant, increase in extract was observed in the mixture[3].

Exploitation of mixtures

Track record Variety mixtures have been extensively used in low input agriculture, and to a lesser extent in high input situations. Land races are locally grown selections of crops, which are heterogeneous and normally contain a mixture of resistance genes that have proved both durable, sufficiently limiting, or tolerant of the prevalent diseases. Land races rarely achieve anywhere near the yield possible with modern varieties, but the latter are dependent upon high fertiliser and, when resistance breaks down, pesticide inputs.

Figure 3 Assessing eveness of barley cell wall modification following malting using a fluorescent dye.

By contrast, the resistance of land races remains stable and they may be better at exploiting lower soil fertility. The essential feature that leads to these desirable features is not the low harvest index, but diversity, and this can be introduced and manipulated to optimise its effectiveness using mixtures of agronomically more useful varieties with high harvest index.

Cereal variety mixtures are grown extensively in several countries where they have been supported by research, notably the USA, Poland, Denmark and Switzerland. Barley mixtures have achieved high malting quality and are used for beer production in some European countries. The biggest problem is not achieving good quality, yield or disease resistance, but convincing end-users that there are not only few, if any, disadvantages to using mixtures, but also many advantages. Unfortunately one of the disadvantages is self-imposed by the end-users. Legislation restricts the sale of mixed seed and maltsters are prevented from trading in mixed varieties by their own rules.

In Switzerland, the 'Extenso' scheme was introduced to reduce pesticide inputs. Farmers are paid a subsidy to grow crops under a low fertiliser, no pesticide regime. Given this restriction farmers rapidly adopted variety mixtures, primarily for disease control but they quickly found the other benefits. Not least of these benefits was a price premium they received as consumers demanded products produced under such conditions. The beneficial implications of growing mixtures may extend beyond the rationale for their use once the less direct economic implications are considered.

Development

Modelling Many mathematical models have been developed to explain the host-pathogen interactions which take place within mixtures resulting in reduced disease levels. These have led to new strategies for optimising mixture composition. There is still much that is not understood about these pathological interactions, and the nature and contribution of the induced resistance component is the subject of current research by SCRI and BioSS. Yield and yield-loss modelling is yet more complex and the integration of yield, yield-loss and disease progress modelling in mixtures is necessary to disect out the effects and optimise these complex interactions. The stochastic modelling techniques we are using to understand induced resistance are likely to be the best route into this intractable jungle.

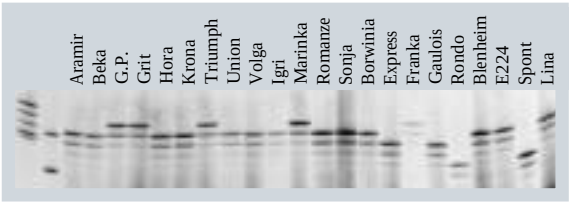

Figure 4 A simple sequence repeat DNA marker showing differences between barley varieties.

Molecular markers Protein characterisation through electrophoresis has been used to distinguish cereal varieties for several years. However, it is generally non-quantitative and not all varieties can be separated unambiguously, so contamination may not be detected. Modern DNA molecular markers, particularly 'microsatellites' or Simple Sequence Repeats (SSRs), are able both to distinguish all varieties[4], and to quantify any contamination. Using only four SSRs, it has been possible to discriminate between all varieties so far tested (Fig. 4). SSRs can therefore be used to identify or verify the composition of a mixture, overcoming a major objection of maltsters that they need to be able to be sure that a grain sample is of the specified composition. The tests are not yet rapid enough for use on grain samples as they arrive in lorries at the maltings, but such technological improvements are likely to be forthcoming. In addition, as DNA is unaffected by the malting process, molecular markers are equally applicable to verifying malt composition.

Perhaps the most important observation from our work on malting quality mixtures is that they were obtained from mixtures of available malting quality cultivars. No attempt was made to optimise the mixtures. Amongst the mixtures were some where the malting quality exceeded the mean of the components. In particular, these were related varieties deriving their quality component *via* a similar pedigree. It may therefore be possible to select components for mixtures that will have consistently better malting quality than any of the component varieties. If these criteria are brought together with optimum combinations for disease reduction and yield enhancement, substantial gains could be achieved very quickly. If combining ability for these attributes were included as criteria in breeding programmes, the gains could be still greater. All these characteristics are potential targets for manipulation using molecular markers closely linked to the desired traits.

Molecular markers are increasingly being used in breeding programmes. The marker is frequently not in the gene responsible for the character being selected, but close by. Many of the genes which contribute to quantitative characters, such as malting quality parameters, can be mapped as quantitative trait loci (QTL) and nearby markers can be used to select for these in breeding[5]. For example, there are markers for six loci controlling fermentability, the factor that determines how much of the malt extract can be fermented into alcohol[6]. Markers could similarly be used for marker-assisted component selection for designing mixtures, as favourable components may combine in a similar manner to favourable alleles in a single cultivar and could explain the high extract levels in certain mixtures.

Potential exploitation in Scotland and the UK In addition to research data, there is need to provide evidence of the commercial value of mixtures, to overcome customer resistance. Molecular markers are likely to be adopted as determinants of varietal purity and extension to mixtures will be a logical extension in a research context. Demonstration of commercial potential, however, requires a crop that is grown over a fairly extensive area, but in which varietal purity is not essential. The world record wheat yield of 13.99 tonnes per hectare was achieved with an equal proportion mixture of the varieties Virtue, Mardler and Husler grown on Mr Gordon Rennie's farm at Clifton Mains in Midlothian in 1981, demonstrating their practical agronomic advantage. Winter wheat is grown in Scotland largely as a source of starch for conversion to alcohol in grain distilling. Unlike bread wheat, high protein levels are undesirable, so distilling wheat is suited to low input systems. The choice of wheat in preference to maize is made by distillers on purely economic grounds, as there is no advantage in alcohol yield, so the optimum raw material may be defined as that which gives the highest spirit yield per unit cost. Although some varieties have been preferred over others, the reasons for this are not clear, so this would seem to be an area where mixtures could be exploited successfully.

In the longer term, benefits of malting barley mixtures of closely related cultivars could offer a means of obtaining malts with particular specifications, not met by most commercially available genotypes. At present, for example, only a few cultivars have low levels of glycosidic nitriles (GN), which undergo a series of chemical reactions during fermentation and distillation, leading to traces of an undesirable component of Scotch whiskies. Tighter regulation or the absence of suitable new cultivars could make distillers seek to

extend the commercial lifespan of some existing cultivars. Varietal mixtures could offer a means of achieving this whilst maintaining profitability, especially as all low GN genotypes can be traced back to a common ancestor, so may demonstrate some similarity in malting behaviour. High diastase varieties are also required for grain whisky distilling. It may be possible to obtain sufficient expression of such characteristics from certain components of a mixture, whilst still maintaining high expression of other desirable characteristics from other components.

Mixtures - the answer to arable farming needs?

Mixtures may not be the full answer for farming needs, but could make a very significant contribution, which is being neglected for the wrong reasons. Mixtures have tended to get consigned to the 'alternative technology' box along with 'organic' agriculture and other 'environmentally friendly' or 'politically green' approaches. Mixtures should be regarded as an approach based on sound scientific principles applicable to many agricultural situations. There are many benefits in their use in low input and 'organic' situations where there are a lack of alternative approaches for controlling disease. Their potential and economic impact is likely to be far greater, however, in mainstream agriculture, where benefits from using the best products of modern breeding programmes and crop production techniques can be further enhanced both in their direct yield response and reliability. Biodiversity provides insurance against unforseen environmental effects. Variety mixtures is an approach that builds this protection into agricultural practice rather than keeping it in store for use in the event of disaster. Mixtures do not remove the requirement for pesticides but may enhance their effectiveness and reduce the level of active ingredient required for reliable effect.

These benefits are achievable with existing varieties and agrochemicals. Available molecular biological tools will enable verification of mixture composition to overcome end-users objections, which can then lead to removal of the legislative hurdles to exploitation. Once routine use in agriculture is established, the incentive will be present to use molecular biological tools in selecting components for mixtures, and for breeding varieties suitable for exploitation in mixtures. Whilst genetic manipulation offers the potential for major advances in disease resistance and yield in many crops, mixtures offer the opportunity to achieve further major improvements in exploiting both new and existing crop varieties. Not least of these is stability of yield and quality, an increasingly important criterion where margins are being squeezed.

In summary

Variety mixtures offer a fast method to exploit all the benefits of modern research, breeding, and agronomic advances whilst providing increased insurance and stability. They offer increased yield, reduced inputs, particularly of pesticides, and improved quality. With relatively low levels of development funding, considerable economic advantage could be achieved. Investment in exploitation of molecular methods and modelling studies would enable much more of the potential of biodiversity to be unlocked and optimised for use in mainstream agriculture. Compared with many other 'environmentally friendly' approaches, use of variety mixtures is likely to have a far greater beneficial effect on the environment as it could be readily adopted for use over a large proportion of cereal growing areas of the world. Perhaps most importantly for its prospects for adoption, it is a method of production that benefits the farmer who will receive direct economic benefit from increased yields and reduced pesticide inputs.

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Organic farming: science and belief

D.K.L. MacKerron, J.M. Duncan, J.R. Hillman, G.R. Mackay, D.J. Robinson, D.L. Trudgill, & R.J. Wheatley

What is 'organic' farming?

The principle There is no single definition for 'organic' farming as the term applies to a movement rather than to a single policy. However, a general feeling for what is 'organic' and what is not can be obtained from the aims of the most prominent associations or organisations within the organic movement. Within the UK, these include the Soil Association. The Soil Association exists "to research, develop and promote sustainable relationships between the soil, plants, animals, people and the biosphere, in order to produce healthy food and other products while protecting and enhancing the environment".

> *"Demand for organic produce is strong and still growing. We are determined that consumers should be able to buy organic produce if they want to. We must remember that conventional systems also produce good quality food that is safe to eat in the quantities consumers demand"*
>
> Mr N Brown, Agriculture Minister, 12 April, 1999 launching a new Organic Farming Scheme.

The unifying principle behind most advocates of organic farming and gardening is the belief that the health of the soil is paramount for life and is the only long-term, sustainable way of cultivating land and creating a secure future for the world.

It would seem to be unreasonable to oppose these broad objectives and so the Agriculture Departments in the UK have accepted that "well practised organic farming is among the options available for environmentally friendly and sustainable production"[1], and that "Organic farming brings benefits to the environment and the consumer through reductions in the use of chemicals and production of food in a healthy natural environment"[2]. The government's Organic Farming Scheme is designed to help encourage an expansion of organic production by providing financial help to farmers and growers when converting to organic methods, a process that lasts for two or more years. Thereafter, produce can be marketed as 'organic'.

Later in this article we will consider whether all of the subsequent implementations of these principles are necessarily so environmentally friendly.

The justification The Soil Association was founded by people who perceived that intensive agricultural systems led to loss of soil through erosion and depletion, decreased the nutritional quality of food, exploited animals in intensive units, and had deleterious effects on the countryside and wildlife. The Soil Association assembled a view of how the best traditions in land and crop management could be combined, and the system of husbandry that they promulgated has since become known as organic farming. The standards that they compiled, together with additions over 25 years, are now used to define the organic system. About 80% of UK organic food is certified by the Soil Association.

Currently, the demand for organic food exceeds the supply from UK sources so that about 70% of organic food is imported from continental European and US sources and organic produce currently attracts premium prices. Organic land in the UK is still less than 0.5% of the agricultural area compared with about 4% in Germany, 5% in Denmark and 8% in Austria, for example.

We have to be clear about which arguments support organic farming on scientific and environmental grounds and which support it on economic grounds. It would seem to be a basic tenet of a free market that nothing that is in adequate supply will attract a premium in price. Note that with those levels of production on the continent, producers there are willing and able to export to the UK at British prices. This could suggest that a similar level of organic farming in the UK would satisfy the market and remove the need for premium prices. Proponents of organic farming should consider, therefore, the possible future economics of organic production if the supply were to increase to meet demand and prices fell to world levels for each commodity. If there are economic benefits then the system will need no support, science or no; and there is growing evidence from within the movement that farmers can reduce external inputs significantly without losing on gross margins, as variable costs are reduced as well as yields. Generally the loss in yield per hectare is 5 - 10 per cent for crops (Table 1) and 10 - 20 per cent for livestock[3]. There are more recent reports of profitability. For example, the report on MAFF project OF0112[4] stated "Gross margins

Enterprise - description	Crop yields	Pesticide use	Fertilizer use	Gross margins
Integrated crop rotations of cereals and legumes, W England	89 - 92%	10 - 81%	69%	103 - 116%
Environmentally benign sugar beet system, E England	78%	nd	nd	102%
Alternative rotation and low input system, W England	S beans 88% Oats 105%	0 - 50% 50%	nd 50%	96% 112%
Standard rotation, low input approach, S England	99.6%	Insecticide 0% Fungicide 50%	nd nd	101% 101%
Reduced fungicide on spring barley, Scotland	102%	12 - 25%	nd	126 - 152%
Supervised low input pesticides on wheat and rape, E England	Wheat 88% OSR 105%	15 - 54% 0 - 96%	nd nd	105% 139%
Integrated farm, the Netherlands	94%	26 - 50%	26%	120%
Biodynamic farms, Baden Wurtemburg, Germany	75 - 91%	zero	zero	80 - 101%
8 biodynamic farms, Switzerland	95 - 100%	zero	zero	100%
Individual organic and 'semi organic' farms, data for wheat, UK	53 - 114%	zero	zero	57 - 138%
58 organic farms, 1986 - 91, Switzerland	73%	P 6%	22%	83%
200 whole mixed farms, 1986 - 91, Germany	Wheat 66% Rye 67% Potato 61%	P 6%	3%	105%
Lautenbach integrated and conventional farms, Germany	99 - 105%	zero	75%	104 - 109%

From Table 7.2 p209. In, *Regenerating Agriculture* by J.N Pretty, Earthscan, London, 1995.

Table 1 Economic indicators for performance of crop components and complete farms of 'sustainable' agriculture in UK and continental Europe as proportion (%) of conventional.

following conversion have been consistently greater than for conventional crops'. Again, SAC in their evidence to the House of Lords[5] stated 'Using SAC's own financial modelling packages, .. even for these all-arable farms, a change to organic production leads to a significant improvement in profitability'.

On the other hand, if the environmental concerns are uppermost then economics are secondary and organic farming may need central support but, in that case, the environmental benefits must be testable and proven.

Encouragement of organic farming EU Member States have promoted organic farming in a variety of ways, principally by subsidies for organic farmers via schemes under Regulation 2078/92. The details of schemes vary widely, most being open to all organic farmers but some only to those converting to organic production. Details of the provisions in the UK can be found at the MAFF and SERAD web sites. Information on the variety of schemes applied within the EU is outlined in a House of Lords Report[6]. Support for converting and continuing organic production from the Community and Member States is estimated to have amounted to over 260 million ecu in 1997. Organic farmers are of course also eligible for support under the various CAP regimes in the same way as other farmers. For British farmers, aid is available through the Organic Farming Scheme or its equivalents in Scotland, Wales, and Northern Ireland. For land eligible for the Arable Area Payments Scheme (AAPS), payments will total £450 plus £600 per farm over the first 3 years[6].

Organic farming is also supported in a variety of other ways. In England, for example, MAFF fund an Organic Conversion Information Service (OCIS), which is run by the Elm Farm Research Centre. Similar schemes exist in Scotland, Wales and Northern Ireland[6]. *Equivalent advice for farmers wishing to use lower inputs in conventional agriculture is neither readily available nor free.*

The Government also provides research and development funding for organic farming: the MAFF organic research and development budget for 1999-2000 is £2.1 million. MAFF marketing grants totalling £1.3 million have also been given out to organic groups[6].

The regulation By April 1999, 60,000 ha of agricultural land in the UK had gained full 'organic' status and another 180,000 ha was undergoing the 2-year conversion together making 1.3% of the total agricul-

tural area. Forty-four percent of that land is classed as unimproved grazing, mainly used for extensive rearing of livestock.

ORGANIC STANDARDS Minimum requirements are set by the EU, the United Kingdom Register of Organic Food Standards (UKROFS, the government control body responsible for implementing the EU regulations in the UK) and the International Federation of Organic Agricultural Movements (IFOAM) and the standards are reviewed regularly.

The standards cover all aspects of organic production during the two-or-more-year conversion period and thereafter, from crop rotations, through management practices for control of weeds, pests and diseases and maintenance of soil fertility, to livestock management[7]. Many of the recommended practices are clearly beneficial. For example, soil management must ensure: a regular input of organic residues in the form of manures and plant remains to maintain the level of humus, biological activity and plant nutrients; a level of microbial activity sufficient to initiate the decay of organic materials … into simple nutrient salts capable of being absorbed by plant roots; and conditions conducive to the continual activity of earthworms and the stabilisation of soil structure. The value of these practices would not be contested; but note that "simple nutrient salts" principally means ammonium and nitrate ions. However, the features that really characterise the organic system are the avoidance of soluble mineral salts as applied fertilizers and the prohibition of agro-chemical biocides. In effect, they forbid a range of practices that are otherwise widely adopted throughout the developed world. The 'soluble mineral salts' that are forbidden are generally the identical 'simple nutrient salts' that are recommended in the organic system when released in uncontrolled quantities by the mineralization of organic matter.

There is one other characteristic that has been included more recently, and that is the attitude towards genetically modified organisms (GMO) and that will be considered later.

The claims and independent assessments

There can be little argument that the adoption of organic farming practices will lead to a number of benefits. So, a project funded by MAFF[4] found that numbers of earthworms increased markedly, and stability of soil aggregates increased giving a more freely draining soil with better structure that needed less work to cultivate it. However, these benefits were associated with an altered and better crop rotation. Almost certainly, it was that rotation rather than the low inputs *per se* which led to the improvements. Data in Table 2, from an independent study that did not involve low inputs, confirm this. Equally, it is clear that the elimination of agro-chemicals in arable areas will lead to increased populations of insects and other biota. What is less clear is whether the changes will be beneficial to people in any real way.

> "*…no other farming systems show such consistent benefits for wildlife.*
> *…crops are habitat just as surely as heather moorland, … "*
>
> MSP Sarah Boyack, Labour Minister for Transport and Environment 2 October 1999.

Treatment** :	1	2	3	4
Soil Organic Matter	2.32	2.16	2.08	2.01
Relative %	100	93	90	87
Instability	1.23	1.25	1.33	1.39
Relative %	100	102	108	113
Crust formation %	83.0	88.4	90.6	91.9
Relative %	100	107	109	111

** Initial soil organic matter = 2.08%

Treatments

1 = 40 t ha^{-1} farmyard manure every 3 years + green manure every 3 years

2 = Input of all crop residues + green manure every 3 years

3 = Green manure every 3 years

4 = Blank, no supplementary input of organic material except roots and stubble

Table 2 Influence of long-term inputs (10 years) of different organic materials on soil organic matter, aggregate instability, and crust formation in silt loam soils[8].

Pesticides SCARAB (MAFF Project PS0401), which was primarily driven by the need to make in-depth observations on the ecological effects of pesticides, found few long-term direct effects of pesticides on non-target insect or spider populations even when they were used at full, manufacturers' recommended rates. However, there were some non-target insects (including ground beetles) and spiders that suffered significant short-term reductions following individual applications of certain pesticides. There was no apparent evidence of long-term trends within any individual species of earthworm. These findings would seem to suggest that the ecological benefits of eliminating pesticides may be more apparent than real. On the other hand, the TALISMAN project (MAFF Project

PS0402) showed that reduced (but positive) use of insecticides improved gross margins on average by 1%. In over 90% of cases there were no significant economic losses, even when certain insecticides were omitted altogether, which suggests that the normal usage in conventional agriculture is higher than it needs to be. It is almost certain that a more flexible approach to reduced use of pesticides could achieve the treble aims of lowering chemical inputs, not damaging the environment, and maintaining profitability.

Quality of food A survey by MORI in June 1999 showed that one third of the public had bought organic food in the previous three months; over half of them because they believed it to be safe and healthy. This attitude is, presumably, based on the perception that hazards in foods derive from agrochemical additives, whereas microbes, not chemicals, are the major source of foodborne illnesses[9]

There are many claims made of organic food, most unsubstantiated and many unwarranted. For example, common claims include[10]: "Organic food is better for you." Has that been proven? And, if so, in what way? We have not found consistent and valid reports of differences in the mineral contents of organic and conventional foods. An early report[11] of differences in mineral content between vegetables grown on widely differing soil types has commonly been misquoted as evidence for the benefits of organic methods. Some reports on qualitative differences, such as flavour in potatoes, can be attributed to differences in dry matter concentration and can be associated with the growing conditions, principally the supplies of water and nitrogen. There, the preferred conditions are as easily provided in conventional culture as in any other. There are many factors, environmental and cultural, that influence the nutritional composition of produce and that are not unique to either cultural system[12]. It is, at best, confusing to try to credit those effects to organic cultivation. Other assertions include that organic food tastes good, is nutritious and is produced without chemical pesticides or synthetic fertilisers. Leaving aside the question of pesticides, there is no difference between the protein and other nitrogen in conventionally and organically grown food. As regards nutrition, there is nothing wrong with synthetic fertilisers. The House of Lords Select Committee[13] found no evidence for or against the safety of organic products. Conventional and organic are equally safe. Further, the evidence given to them by the British Nutrition Foundation was that "the nutritional value of organic crops is likely to be the same as that of conventionally grown crops"[13]. The Select Committee emphasised that the organic label certifies that a product has been produced in a particular way; it is not that it has certain desirable qualities. Organic standards are based on the method of production, not on the characteristics of the finished product. They recognised that, in purchasing organic food, consumers may be expressing preferences other than the content of the food itself.

Consumers should beware of mission-directed disinformation from pressure groups. That was the assessment by the American Council on Science and Health, a consortium of over 250 leading scientists and physicians, on a report on pesticide residues on fruits and vegetables by a concern called the Environmental Working Group (EWG)[14]. In that report the EWG had excited fears over residues that led to the withdrawal of fruit from school menus. Yet the thresholds used were the official 'reference doses', only 1% of the level that should trigger questions about the source, and a smaller fraction still of the permissible dose.

Organic food is guaranteed free from genetic 'tampering'[7]. This was not part of the original principles and opposition to genetic modification appears to be a hitchhiker or a stowaway cadging a ride on a respectable movement. The matter will be considered later.

The Soil Association has adopted six criteria for the assessment of the quality of food - Sensual, Authenticity, Functional, Nutritional, Biological, and Ethical - to define a holistic approach. Such criteria defy the inclusion of material that is not *a priori* part of the organic system.

Nitrate in water Water leached from organic farms has been reported usually to contain less nitrate than the EC nitrate limit of 50 mg / litre although, in MAFF projects NT1313 and OF01410, the limit was sometimes exceeded[4]. That claim is not entirely borne out by the data from those projects, Table 3.

A similar statement could be made about conventional farms. Indeed, in those projects, comparison with conventional arable and grassland farms over the same period showed that nitrate losses from organic and ordinary arable fields were similar, although losses from conventional, intensively managed leys, including the ploughing out stage, were said to be higher than from organic leys. Neither details of the conven-

Year	Crop	Mean (mg nitrate/ litre)	Range
1993/94	Ley Arable1 Arable2+	20 68 26	0.4 - 25 34 - 112 8 - 58
1994/5	Ley Arable1 Arable2+	23 84 56	0.4 - 138 12 - 126 7 - 102
1995/6	Ley Arable1 Arable2+	45 105 113	1 - 108 8 - 201 44 - 368

Arable 1, first after ley; Arable 2+, all subsequent arable crops after the first.

Table 3 Nitrate in soil water from the organic farms[4].

tional rotations used for comparison nor the data on nitrate leached from them were given in the published report[4]. However, nitrate leaching in the two systems was said to be highly variable and the data shown (Table 3) is most revealing. Had the data come from a conventional system, one could well have inferred that there were strong arguments for reducing the inputs of fertilizer nitrogen. But that cannot be said of an organic system. Instead, we are left with the conclusion that the supply of nitrate was not at all controlled in the organic system and was not at all well matched to the demands of the crops being grown. It is striking that nitrate in the soil water was not highest after the ley, the 'fertility building phase', but after one or two or more arable crops. Clearly, the more extreme losses of nitrate should be avoided. Whether this can be achieved within the presently defined organic system has still to be proven. The balance of environmental advantages and disadvantages in the organic system is not clear. The House of Lords Select Committee on the European Communities did not accept that organic farming is the only way to achieve environmental and other benefits[15].

Economic aspects The economics of the organic approach are not really a concern of this review of organic farming but they cannot be ignored. It was reported[4] for MAFF Project OF0112 that gross margins, following conversion, had been consistently greater than for conventional crops and, even when projected over a full rotation and to a farm scale, the organic rotation had been significantly more profitable than the conventional rotation every year. The CWS study over 8 years concluded that, 'Both the mixed and all-arable organic systems were only slightly less profitable than the conventional farm data comparisons'[16].

Currently, organic farming in the UK is predominantly on mixed and livestock farms in the West and North that produce animal manure. If the demand for organic arable and vegetable products continues to increase then it is likely that more farms that are currently all-arable will be considered for conversion[4]. These will face particular problems in maintaining soil fertility unless they become mixed farms with all the investment in infrastructure that would entail. The 'balanced' rotation envisaged for organic farms typically includes 2 or 3 years of grass-clover ley in a 5- or 6-year rotation. If the farm does not carry livestock, that land is, essentially, set-aside. True, none of the costs of producing a crop on that land are incurred but neither does it yield. It does not contribute to the income of the farm in those years and so the production from such a farm, as a whole, is halved. Suddenly the economics of the operation seem much less favourable. The House of Lords Select Committee on the European Communities rejected the idea of sustained subsidies for organic farming[15] but did commend continued financial support for conversion to organic farming[17].

Other environmental issues Organic farming is claimed to be better for the countryside. For example, birds and other wildlife are a valued part of organic farming. It is a matter of record that less intensive application of conventional methods achieves the same results. Lapwings thrive on permanent pasture. So, increases in their populations, claimed for organic farms, reflect the balance between extensive and intensive agriculture, not an effect of organic farming *per se*. Several witnesses to the House of Lords Select Committee held that increases in biodiversity result from identifiable changes in management that could be implemented on conventional farms[18]. Recommendations on the treatment of hedges similarly can be accommodated within conventional systems and often are. It is a mistake to classify 'conventional' farms uniformly or even simply to divide them between all-arable, mixed, and livestock farms. There is a range of levels of environmental awareness in their management. Conventional farms can be, and often are, managed in ways that provide the benefits to wild life claimed by the organic movement[18].

Organic standards through the EU and elsewhere The standards set by UKROFS require that the crop rotation (and therefore, crop nutrient supply) be based on a balance between crops that build fertility (i.e. legumes, normally grass/clover mixtures) and those that exploit it. The Soil Association declares in

its standards[7] that 'Brought-in manures or plant wastes from non-organic sources must not form the basis of a manurial programme, but should be adjuncts.' Yet there will be little of such materials available from organic sources as these are intended to run 'in balance' and should not be producing exportable wastes. Public controls on what may be fed to livestock and the protection of the public from residues in meat should mean that dung from a beast fed on an 'organic' farm is much like that from one fed on a conventional farm. Certainly, the differences between types of animal manure - poultry, pigs, and cattle - are far greater than those between manures from the two systems[19]. There seems to be little justification for this stance other than refusal to connive at another's spreading the resources of his farm. It is not clear that other European countries share these scruples. In the Netherlands, there is a much greater concern with the rational use of manure that is recognisably in surplus (at least locally) and that each farm should have a balance of inputs and outputs of each of the principal nutrients. While Dutch agriculture is being restructured, it is a requirement that animal producers should either have the land to take up the waste or have a contract from another farmer to use it. The sale and export of manures is encouraged. The USDA, in trying to define organic production, specifically includes off-farm organic wastes but whether or not those wastes are 'organic' in the sense used by the organic farming movement is unclear. The contrast between what the Dutch see as an evolution towards ecological agriculture[20] and the strict self-sufficiency of the organic system operated in the UK is striking.

The problems

Sustainability Although it is relatively easy to describe goals for a more sustainable agriculture, it is more problematic to define sustainability which is a complex and contested concept. To some, it implies persistence and the capacity for something to continue for a long time. To others, it implies resilience and the ability to recover from imposed difficulties. Applied to the environment, it involves actions that do not damage or degrade natural resources. Others see it to mean that developmental activities simply take account of the environment. In any discussion of sustainability, it is important to clarify what is being sustained, for how long, for whose benefit and at whose cost, over what area, and measured by what criteria. Answering these questions is difficult as it means assessing and trading off values and beliefs[3].

'Attempts to define sustainability miss the point that, like beauty, sustainability is in the eye of the beholder ..'

A. Campbell [21]

What do we mean by sustainable? All the food and other products taken from the land represent an abstraction of resources. Unless these are replaced, that land will become depleted and infertile. So, both organic and conventional agriculture must look to what resources are drawn from the land and how these are to be replaced. The principal commodities taken from the land are water, carbon and energy. These three are renewable. The water is replaced by rain and the amount of water that is taken off in agricultural products is a very small fraction of the total throughput in the system. Energy from the sun is fixed during photosynthesis in which growing plants assimilate carbon from the atmosphere. The ability of plants to fix carbon and energy is dependent on the fertility of the soil and its physical properties. The organic matter in the soil influences these and so it is a legitimate concern that an adequate proportion of the assimilated carbon should be left in the soil. The supply of water as rainfall may be inadequate for maximum crop production, and then irrigation may be considered. Unless the irrigation water is drawn from on-farm reservoirs, filled by winter rain, the practice of irrigation is arguably not strictly sustainable; but this has not yet been considered as an issue by the organic movement.

The main concerns over sustainability must lie in the ability to replace the nitrogen and other mineral elements that are taken from the land in a crop. Nitrogen and some other minerals are replaced from the atmosphere but not at rates that even approximate the rate of abstraction in a crop. So, the fixation of atmospheric nitrogen is in the order of 10 - 80 kg N / ha / y over most crops and 80 - 280 kg N / ha / y over a clover-rich sward. But a well-grown crop of potatoes (60 t / ha) takes 160 kg N / ha off the field in the tubers and a 5 t / ha crop of spring barley would remove about 85 kg N / ha in the grain and perhaps 40 kg N / ha in the straw. From published figures of the mineral composition of potato tubers, it is a simple matter to calculate that a crop of 60 t / ha of potatoes removes 29, 338, 12, 4, and 5 kg per hectare of P, K, Mg, Ca, and Na, respectively, in the tubers. These minerals are not readily replaced in rainfall (Allen reported[22] average annual deposition in rainfall over seven sites to have

been 0.5, 4.0, 4.1, 11.4, 33 kg / ha of P, K, Mg, Ca, and Na, respectively, and 14 kg N / ha) and the growth of green manures only serves to cycle them.

"Sustainable agriculture should not imply a rejection of conventional practices but the combination of the best opportunities from modern science with a re-adoption of traditional opportunities to conserve resources. The two themes need not be incompatible". (Paraphrased from Pretty[3]).

Crop nutrition Is organic cultivation an environmentally friendly and effective agronomic system in which all resources are used effectively or are some wasted? Organic wastes should be incorporated into the soil directly after application. If this is not done then significant quantities of nitrogen can be lost to the atmosphere as ammonium and as nitrous oxide (N_2O) which will not only be losses from the system but are both atmospheric pollutants. In addition, the recommendation[7] to compost manures before using them is a practice that ensures significant loss of nitrogen to the atmosphere as ammonia. It is ecologically unsound. The maximum amount of the nitrogen in animal wastes is retained where slurry is placed directly into soil that has a growing green crop such as pasture. This is the required procedure in the Netherlands, in contrast to the common British practice of spreading slurry or manure on the surface for later incorporation.

When fresh manures or harvest residues are ploughed in, a proportion of the nitrogen content is readily mineralized to nitrate and this occurs within only a few weeks[23]. The remainder becomes available slowly over a period of years. The nitrate is immediately available for uptake by a crop, immobilization, denitrification, or it may be leached. Depending on the time of incorporation, more or less of the nitrogen in the manure can be available to the following crop.

How can one ensure that the nutrients are available to the crop at the correct time, and that mis-timing does not cause either environmental damage or reduction in crop yield and quality? The timing of tillage and the application of manures are central to the phased release of mineralised nitrogen. The rate of mineralization of organic matter in the soil is enhanced by cultivation, which should be timed so that the most nitrogen is provided shortly after emergence, when it is required by the crop. An inadequate supply of nitrogen during canopy expansion results in poorer interception of light and lower yield. Late mineralization of organic nitrogen to available forms, as happens after the harvesting of root crops or autumn ploughing, can result in losses through leaching.

The logistics of controlling the time and amount of ammonium-nitrogen released from the complexity of soil organic matter, and its subsequent conversion to the mobile nitrate-nitrogen, is not a trivial task and is a problem that is common to all husbandry systems.

It is likely, therefore, that computer-based decision support systems will play an increasing part in land management. For example, a package called MANNER (MANure Nitrogen Evaluation Routine) has been developed by ADAS with support from MAFF (Project No. NT1423) that uses a few simple inputs to characterise the organic matter being added to the soil. It then estimates losses by volatilization of ammonia, and leaching of nitrate, then the mineralization of manure N and, finally, the amount of manure N that will be available to the crop. Packages such as this will allow improved use of organic manures and a rational way to combine organic and bagged fertilizer. It is naïve to consider that a system that relies on the use of materials that break down slowly will allow better control of supply than one that uses a mixture of slow turnover and rapid correction.

Pests All crops can be attacked by pests and diseases so that the general expectation is for crops to suffer where chemical controls are removed. However, other management options may be available depending upon the crop and the pest or disease. Each has to be considered as a special case. Only a few will be considered here.

NEMATODES Nematicides are amongst the most toxic pesticides used and, because they have to be applied to soil, their rates of application are also high. In practice, a nematicide such as aldicarb is usually safe for people because of its restricted availability, the methods for its incorporation that prevent people coming in contact with the product, and because it rapidly degrades in most soils to harmless compounds. However, it is toxic to other soil animals, e.g. earthworms, and so has undesirable effects. Other soil nematicides or fumigants such as methyl bromide are even more undesirable.

So what are the options available to limit the effects of nematodes?

Although very damaging in some countries and although they occur in c. 50% of UK cereal fields, cereal cyst nematodes only occasionally cause damage

in the UK. Here, two fungi control it, one of which is almost specific and the other is a facultative egg parasite. These biological control agents increase and become suppressive following repeated cereal cropping. It seems likely that many other potentially damaging, mainly ectoparasitic, soil nematodes are kept in check by a whole complex of parasites and predators about which we know very little.

That is an example of a biological control that is effective without human intervention. However, in general it is extremely difficult to make biological control work in soil because of the complexity of the soil environment. There are problems in introducing the control agent, and once introduced it has to compete with the multitude of other organisms present. It has to persist and increase, and it has to be effective. Ideally, it needs to be specific to the pathogen against which it is targeted, and it needs to be safe to use. The case of control of the cereal cyst nematode is the exception rather than the rule.

Management by control of the rotation works for some nematodes, such as cyst nematodes with narrow host ranges, but not for others. This method of control requires a knowledge of principal and alternative hosts, rates of population decline when non-hosts are grown and of increase when hosts are grown. For other nematodes with wide host ranges, the position is even more difficult.

Some work has been done at SCRI on a specific bacterial parasite *(Pasteuria penetrans)* of the root-knot nematode *(Meloidogyne)* in an EU-funded project. Some soil types are unsuitable for its deployment but, even in those that are suitable, it is rarely suppressive. However, massive increases in soil populations of the bacterium and impressive suppression of the nematode have been observed where the gene pool of an indigenous strain has been enhanced by the introduction of an exotic one. Understanding the genetics and dynamics of such systems is not trivial, but it is possible that they could be developed for a few defined cases.

With species such as potato cyst nematode (PCN), where egg hatching is involved, trap cropping can be helpful but it requires a thorough knowledge of the biology of the pest, and may not be possible in many situations. It is, in effect, the mechanism that allows short rotations on land used for the production of early potatoes, where the crop is harvested before the nematode has time to complete its life-cycle.

In some circumstances, physical control can work. At mid- to low-latitudes the most promising method is solarization but that is just not an option in the UK. Flooding, where possible, could be used to reduce numbers of pests but would also have deleterious effects on the beneficial biota and on soil structure.

The remaining choices are the use of resistant varieties of crops and the use of 'integrated pest management'. Resistance is generally seen as the long-term solution to pests and diseases. However, the goal of producing a variety that has the nutritional and culinary qualities that the market wants and that has resistance to all the threatened pests and diseases is unlikely to be attained in the foreseeable future. Certainly, not by conventional breeding. Producing resistance to even one pest is a major task although the rewards are considerable where it is achieved. It is the most cost-effective solution and involves no use of agro-chemicals. The potato variety Maris Piper, and similar ones, resistant to *Globodera rostochiensis*, the yellow PCN, gave enormous benefits to potato farmers in the UK during the 1970s and 1980s. Unfortunately, most farmers ignored the warnings of nematologists and did not integrate that resistance effectively with rotation, use of nematicides and an alternation between susceptible and resistant cultivars. The result has been that the virulent white PCN, *Globodera pallida*, has selectively replaced the yellow form in a very short time.

SCRI is heavily involved in studying sources of resistance in the host, characterising races of the nematodes for components of virulence, and the interaction between host and nematode. Activities to integrate that knowledge in mathematical models indicate that the white species, *G. pallida*, can be almost impossible to control without integrating resistance into a strategy which also includes long rotations and the use of granular nematicides.

Some level of integrated control is practised by almost all farmers, organic or not, because most use rotation. However, for pathogens such as PCN, proper control is not possible without the integration of two or more methods unless rotations are to be extremely long (>12 years). This will be discussed further under 'Opportunities'.

APHIDS AND OTHER INSECTS Aphids are vectors of viral diseases. In seed crops, therefore, the threat that they pose to the value of the crop is out of all proportion to their numbers. Even in the north of England

and in Scotland where climatic conditions delay the appearance of numbers of aphids, growers depend on the use of chemical sprays to prevent loss of seed crops of potato. Aphids can form serious infestations in crops of both peas and beans that may even lead to the direct loss of the crop[16]. Yet there is no effective treatment available in the organic system. An example of the potential for a low input, biological control in this area is described under 'Opportunities'.

SLUGS Slugs are favoured by weedy conditions, a likely circumstance in organic culture. Some varieties of potato are more resistant to slugs than others but, in all cases, slugs can make large amounts of tubers unsaleable. There have been cases reported of organic cereal crops being lost to slugs[16]. Again, there is no effective treatment for field crops in the arable system.

Disease Most of the general observations on pests also apply to fungal and bacterial diseases of crops. Approval for the use of Bordeaux mixture in any crops will be withdrawn by the EU from 2002, so that the organic system will be left with few means other than genetic resistance to combat disease. This is discussed further in the next section. Zwankhuisen et al.[24] investigated the origins of outbreaks of late blight of potato in the Netherlands. They found that 74% of the early outbreaks were associated with nearby cull heaps, etc. Infected seed tubers and volunteer plants were of minor importance. Later, in mid-growing season of a year favourable to the spread of late blight, they found that infested organic potato fields became a secondary source of infection. In the Netherlands, the foliage of organic crops has to be destroyed by flaming immediately after the first appearance of the disease in order to prevent dispersal to neighbouring fields, to reduce the risk of oospore formation and to avert the infection of seed potatoes. There is a double message in this. One is that organically farmed potatoes are more prone to late blight and significantly increase the infection pressure on neighbouring crops. The other is that the moderate success to date in avoiding late blight in organically farmed potatoes is attributable to their low density - and, possibly, the use of Bordeaux mixture. They are surrounded by protected, clean, conventional crops. In this the organic growers are not unlike those parents who elect not to have their children vaccinated against a disease. They are, in effect, reliant upon the good health and hygiene of their neighbours.

Weed control Weeds pose the greatest challenges on some farms[16]. The need for a number of cash crops in the rotation leads to problems with both annual and perennial weeds. Extending the ley or green manure periods would reduce the weed problem but also the returns. Techniques that are available in an arable rotation include stale seedbed (not quite as stale as practised by conventional farmers), adjustment of sowing times and rates, and mechanical methods but these last are either laborious or expensive in energy.

The contradictions - Environmental Friend or Foe?

Pesticides based on bacteria and viruses offer promising opportunities for selectivity in tackling pests and in reducing pollution[3 - p. 103]. The greatest successes so far have been preparations derived from *Bacillus thuringiensis* (*Bt*)[25]. The bacillus produces a soluble crystalline toxin that paralyses the gut and mouthparts. The toxin is effective against a range of insects, particularly certain lepidoptera, but is harmless to plants and to humans. It is used in a wide range of pesticides and the bacterium itself is used for pest control in organic systems[25]. The toxin of *Bt* is produced by a single gene which has now been cloned and inserted into non-pathogenic bacteria that colonise plant roots, and also directly into some crop plants such as tomato. The potential for engineering plants to contain their own defensive compounds in this way is considerable. The Soil Association's standards[7] accept *Bt* as a chemical but not the insertion of the *Bt* gene into the plant itself. Yet many crop plants produce localised toxins. For example, potato produces high levels of glycoalkaloids in its leaves as a defence mechanism, but not in the tubers unless they are allowed to 'green'.

In the report on a recent MAFF-funded project (OF0112) it was reported[4] that "The only pesticide applied was Bordeaux mixture. This copper-based fungicide was applied to potatoes up to three times a year….". The justification for the use of Bordeaux mixture - a simple solution of inorganic salts - in 'organic' systems appears to have been that it was hallowed by time. In fact, it is not at all environmentally friendly and copper is toxic at the levels used in Bordeaux mixture - that is why it was devised. Bordeaux mixture is a known molluscicide, it repels slugs and snails and is toxic to earthworms, and its use within the EU will be banned in 2002 - nothing to do with organic farming, rather the environmental concerns of the 'conventional' farming community.

A full account of the use and toxicology of copper sulphate is to be found at the web site: http://pmep.cce.

cornell.edu/profiles/extoxnet/carbaryl-dicrotophos/copper-sulfate-ext.html

Conventional agriculture has reached its present level of production by adopting the use of a battery of agro-chemicals - fungicides, insecticides, nematicides, herbicides - and these are among the products that have been most opposed by the organic farming movement. Traditionally, plant breeding for resistance to diseases has been seen as an important means for agricultural progress and reduced reliance on agro-chemicals. The modern techniques, familiarly called genetic engineering or genetic modification, offer the prospect of introducing resistance to pests and diseases more effectively and more quickly. After taking evidence from a wide range of experts and interest groups, a select committee of the House of Lords reported[26] that, "..biotechnology in general and genetic modification in particular offer great potential benefits to agriculture, industry, consumers and even to the environment. The fruits of the technology should be available to our farmers, manufacturers and consumers. These developments have to be surrounded by an assessment of risk (and, where necessary, its management), ….." Yet the Soil Association, in its evidence[27], described the introduction of genetically modified plants into United Kingdom agriculture as the "most serious threat ever to the objectives and progress of the organic farming movement in developing and introducing viable systems-based approaches to agriculture". This opposition has to be recognised as a contradiction of the underlying principles of the organic movement. No-one objects that there is no food for the caterpillar of the cabbage white butterfly where cereals are grown. Why should they complain if they couldn't feed on a cabbage crop? They may still feed on wild relatives of the cabbage. Where is the ecological benefit of rampant late blight of potato? No other organism feeds on the fungus causing the disease. Genetic engineering would appear to be compatible with the principles of organic farming even if the proponents of the movement do not presently recognise it as such. The relation between genetically modified organisms (GMO) and organic farming will be discussed again in the next section.

The opportunities

Genetic modification For over 70 years, plant breeding and selection for resistance to diseases has been seen as an important means for agricultural progress. This strategy has had some successes but in many cases, for example rice blast in rice and late blight of potato, disease resistance in the crop has been relatively short-lived and the disease organism has mutated or been selected to overcome that resistance. If durable forms of resistance to any of the major diseases could be introduced into otherwise acceptable varieties by genetic modification, the saving in use of agro-chemicals would be immense.

The Soil Association has, however, set its face against the use of genetically modified crops in any form[7], stating "4.303 Prohibited .. 4) Varieties of seed that have been produced using Genetic Engineering". There is no doubt, no equivocation, just straight opposition. In doing this, the Soil Association would appear to be opposing the very means by which many of its ends could be achieved. They should be encouraged to take a more positive role by participating in determining the standards that might be used to test for acceptability and safety of GM products. They should help to establish what is safe. The corollary, of course, would be that once there are agreed standards for testing, material that meets those standards would be acceptable.

At present, the Soil Association and its present allies consider that not only are GM crops unacceptable but also that no pollination of organic crops by pollen from GM crops is tolerable. More recently they have extended this objection to passive contamination with pollen from sexually incompatible species and the aversion extends to the inclusion of such pollen in 'organic' honey. The former point seems to be particularly illogical. Sexual barriers prevent the exchange of genetic information. The latter one can hardly be achievable given the foraging range of bees.

The zero-tolerance approach to pollen is clearly incompatible with the coexistence of GM and organic crops. The only way to achieve it would be to ban GM crops entirely - or to abandon organic farming. There must be other ways. For example, there are established tolerances for other kinds of contamination in organic products. Council Regulation 2092/91 allows 5% of non-organic ingredient in organic products, although a draft regulation proposes reducing this to 1%. Again, ACRE in 1998, considering a case involving organic maize, noted that the purity requirement for Basic Seed (the highest specification for maize seed) is 99.9%. This sets an upper limit on the purity that can be guaranteed for the crop. Therefore, pollination by GM pollen at a level much less than 0.1% could not reasonably be regarded as significant.

If a non-zero tolerance level were to become accept-

able, there would be scientific questions to be answered in determining what that level should be such as how compliance is to be verified, and what isolation or other measures are required to ensure compliance.

Non-zero tolerances are consistent with other aspects of organic farming. What often seem to be arbitrary rules can in many cases be represented as a compromise between what is considered desirable and what is practicable.

Integrated crop management

MANAGING NATURAL ENEMIES - PREDATORS, PARASITES, AND DISEASES The use of natural enemies is commonly referred to as biological control and examples have already been described under 'Nematodes'. That can be further classified as 'classical', in which new or exotic natural enemies are released, and 'augmentation' that relies on improving or supplementing the existing control. There have been considerable efforts over recent years to develop effective biological control programmes[28]. Occasionally the results are spectacular as in the control of *Opuntia* with *Cactoblastis cactorum*. More usually programmes have moderate success and there have been more failures than successes[29]. The principal difficulty is in maintaining the parasite or predator at levels that will keep the pest at an acceptably low level. For this reason, some of the most successful programmes for biological control have been against pests of glasshouse crops where the high level of environmental control coupled with containment and replacement favour the use of natural enemies.

An interesting example can be taken from another environment. Pigeon pea and cotton share a common insect pest *Helicoverpa armigera* that is highly damaging to both crops and that has developed a considerable amount of resistance to pesticides. To tackle the problem, the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), in collaboration with national programmes and other research institutes, has been studying a naturally occurring viral disease, nuclear polyhedrosis virus (NPV). This is specific to *H. armigera*, and has been found to be 95% as effective as chemical insecticides[30], giving low-cost control and an environmentally safe option for subsistence farmers who grow pigeon pea. Pigeon pea is also used as a trap crop, grown with cotton[30].

TECHNOLOGY AND REDUCED INPUTS In the UK, evidence from growers is that if the timing of application is correct for fungicides on cereals, rates can be cut by 50 - 75% and still maintain yields (See Table 1). For example, a recent recommendation is that farmers should inspect their crops regularly and apply quarter rate fungicide mix when 75% of plants show at least one active spot of mildew[31]. Careful monitoring and sequential sampling for pests on brassicas has reduced the need for pesticides by 85%, while maintaining yields. In the soft fruit sector some growers have cut their use of fungicides to 12 - 25% of former levels after adoption of a range of IPM techniques[32]. However, these low-dose approaches do demand that growers monitor their crops intensively. In a sense, the two extremes of organic and conventional agriculture discourage thinking. Either you apply nothing (organic) or you apply the full dose to everything (conventional). A rational approach requires monitoring and decision making. The rewards can be considerable, both financial and environmental.

Patch spraying is a technique that similarly can reduce inputs significantly. Patch spraying needs a combination of regular field monitoring, whether done using modern technological capabilities such as remote sensing coupled with GPS or more simply from direct field walking, and modified spraying systems. The approach is applicable for control of both weeds and pests.

Conclusions

Organic farming has the joint aims of being environmentally sensitive or correct and being sustainable. In order to promulgate those aims, it has developed a set of rules and standards to which its followers must adhere. There is very little derogation. As a result, organic farming does not require best use of the options available, but the best use of the options that have been approved. These options are usually more complex and sometimes less effective than conventional ones. Improvement of the options available to the organic farming movement will require scientific effort - it may also require some re-evaluation of practices by its adherents. Such science will also be relevant to conventional agriculture, because many practices are common to both organic and conventional farming and because the 'conventional' farmer is not inhibited from using 'organic' approaches. So, for example, a conventional farmer may conclude that the 'organic' approach is about right for pesticides and only half right for fertilisers, or whatever.

The philosophy of the proponents of organic farming presents a difficulty to the scientist. The organic movement, generally, resists comparisons between

adjacent and otherwise comparable plots of 'conventional' and 'organic' land, saying that the organic system has to be considered as a whole and that it is inappropriate to break down parts for examination. Yet, without such an analytical approach it is not possible to understand, or even monitor satisfactorily, the processes operating within the system. This philosophical disparity is at least a contributory cause for the lack of valid comparisons between systems in soil fertility, pollutants in soil solution, the value of the food produced, and others.

The organic farming movement presents a challenge to the scientist, who cannot - and would not want to - abandon a scientific approach. We have to accept a philosophy that says, 'I don't want to do particular things.' The challenge for the scientist is to find a way of living beside, and working with, that system, while recognising that not all of its tenets are correct, and while identifying which of them are misguided, and enabling the improved application of those that are correct. Where the organic system does not conflict with science, there is no problem but it is possible to show, for example, that strict adherence to some principles such as sources of fertilizer, could be more harmful to the environment than other ways. The present lower productivity of the system should not present a conflict. It is simply the result of a constraint that the practitioners have put upon themselves in order to achieve a particular effect in their environment. While the intended effect is one that most people agree is desirable, there may be other, more direct means to achieve it. Where a scientist has particular difficulty is with the evangelising statements that the products of the system - food and fibre - are in some way 'better' than those from conventional agriculture.

We would like to be able to argue for a different route in some aspects, between 'organic' and 'conventional, high input' farming. An exemplary issue is the production of varieties that are resistant to pests and diseases - particularly where the adoption of those varieties would lead to the reduction or even elimination of particular chemical inputs. Instead, the pharisaic attitude of many in the organic movement leads to feelings of frustration in some scientists who can see the applicability of their work to the system, yet have it rejected. The products of biotechnology in general and of genetic engineering in particular are among that work.

A recognised major disadvantage of organic farming is the likelihood of reduced output per hectare. The extent of this reduction can be limited by careful rotational planning, strategies for efficient manuring and supply of crop nutrients and by attention to detail in the husbandry practices adopted for control of weeds, pests and diseases. This is another case where a flexible approach to solving problems by, for example, combining modern technology for monitoring and positioning with selective use of agro-chemicals, is capable of transforming crop production while minimising inputs.

A scientific base is essential to make best use of the available measures for production and control of problems in conventional agriculture, and even more so in organic agriculture. Just as the consuming public has a right to a choice in the products that it will use, based on professionally produced information, so the farming public has a right to scientific and technical support for their agricultural systems, which ever is used. But they only have that right where they are prepared to accept, for a time, the validity of its products. All good science accepts that its results may later be proved false. Where an approach to agriculture, or life, rejects the lessons of science *a priori*, then it disqualifies itself for that support. The organic movement should be encouraged to be less defensive of its standards and less prescriptive in their application. It should be encouraged to recognise the contribution towards its wider aims that is offered by modern science. Equally, scientists must recognise the opportunities for interesting science offered by the constraints of minimised inputs and the principle of sustainability. SCRI has a mission in these areas.

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Plant molecular and cell biology

Gordon C. Machray

The synergy afforded by the application of cutting-edge research in molecular biology and in cell biology is apparent in the study of plant science at SCRI. Publication in prestigious journals, top ranking in research assessment exercises, major external contracts, and the generation of intellectual property, all demonstrate the health and vigour which has characterised our efforts in this area. Underpinning these are the talent and enthusiasm of scientific and support staff. We must ensure the consolidation of areas of excellence and explore all opportunities to emulate these successes.

Gene expression and RNA processing Precursor messenger RNA (pre-mRNA) splicing is one level at which gene expression is regulated. The Gene Expression/RNA Processing Group has been studying two inter-related areas of plant splicing: exon scanning and mini-exon splicing. Significant contributions to understanding the process of splicing have been made. Firstly, it has been shown that exons can be defined by interactions between factors assembled at each end of the exon, aiding our knowledge of how splice sites are chosen. Secondly, splicing elements required for correct splicing of a small (nine nucleotide) potato invertase mini-exon have been identified. These sequence elements are also able to promote splicing of heterologous exons of only one nucleotide in length. Splicing is very efficient, reflecting the strength of the splicing signals. Initial experiments suggest that these sequences can act as a splicing enhancer to increase the efficiency of splicing. Whether this will also lead to an increase in levels of gene expression is still to be investigated, but identification of splicing enhancers or expression enhancers will be of value to biotechnologists for the development of improved plant material. The various RNA and protein molecules involved in splicing are often encoded by gene families within the plant genome. We have

developed several novel approaches to the determination of expression profiles of individual members of plant gene families. These have revealed tissue-specific or constitutive expression patterns for individual gene family members, indicating potential uses for the promoters of these genes. Further methods for the subsequent isolation of these unique promoters from large complex genomes, such as tetraploid potato and hexaploid wheat, have yielded promoters eminently suited for biotechnological application for which intellectual property rights have been obtained.

Using these methods, we have confirmed constitutive expression of several genes from the U1A and U2B'' gene families in potato, with one expressed at 65% of the level of the CaMV 35S promoter. Promoters such as this are of increasing importance, given concerns over gene silencing resulting from promoter duplication and the use of promoters derived from viruses or other plant pathogens. Among the best-characterised tissue-specific promoters we have cloned are those for the family of genes encoding cell wall invertases in potato (see pp. 82-85). For each of these four promoters, we have generated a series of transgenic plants expressing a reporter enzyme under the control of the potato promoter, allowing detailed histochemical analysis of expression profiles. One promoter determines gene expression at the axial node in the stem under the axillary bud. Expression is also seen in an analogous region of the root. The axial nodes have major importance in potato - this is where the stolon, which will give rise to the tuber, is initiated, and, in the tuber itself, it is the region which forms the 'eye' from which the sprout will emerge. This promoter may be applied to modulation of development of the plant, or pathogen control strategies. Two promoters govern gene expression in the vascular bundles of the potato stem (there are three major and three minor bundles). One of these is preferentially expressed in the internal phloem of the bundle while the second is preferentially expressed in external phloem. These promoters may find application in insect control strategies, or in the modulation of stem nutritional content. There are early indications that expression of one may also be induced in roots by nematode attack, suggesting a further use in nematicidal strategies. The fourth promoter governs expression in developing pollen cells of the anther. This promoter may have application in pollen ablation for male sterility - we have shown that it is active in potato and tobacco and its utility in gametic transformation protocols is also being assessed.

Plant transformation remains an area of active interest. Cereal transformation using the biolistic approach has been achieved with the generation of transgenic wheat containing constructs bearing genes for coat protein or movement protein from soil-borne wheat mosaic virus. In this EU-funded project, with the goal of engineering resistance to the major viral pathogen of wheat in the under-developed world, both model cultivars and cultivars of importance in Chinese agriculture have been transformed. The genotypic dependency of transformation is a current focus of our work on barley transformation, which has revealed significant differences in regeneration frequencies within the pedigree of cv. Golden Promise, the model cultivar routinely used for transformation. Difficulties in the regeneration phase remain the primary problem in the facile transformation of barley cultivars in current use in UK agriculture.

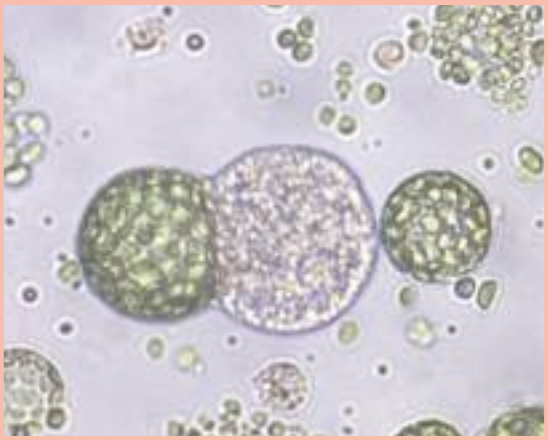

Figure 1 A tobacco-barley heterokaryon resulting from protoplast electrofusion (photograph courtesy of Julie Wardrop).

Transgenic barley (Golden Promise) is also being used in a novel radiation hybrid approach to gene mapping in barley. Radiation hybrid mapping is based on the generation of a panel of hybrid callus lines derived from the fusion of herbicide-resistant barley protoplasts to tobacco protoplasts (see Fig. 1). Selection for herbicide-resistance under conditions otherwise unfavourable to the barley cells, encourages the addition of segments of the barley genome (including the herbicide-resistance gene) to the stable tobacco genome. Analysis of co-transfer of linked markers then allows a physical map of the barley genome to be assembled. This, and other *in vitro* mapping approaches based on physical fragmentation of the barley genome, its dilution into fractions of less than a haploid equivalent, then reamplification of the frac-

tionated sub-genome (HAPPY mapping), should allow the construction of physical maps complementary to barley genetic maps based on molecular markers - an area in which SCRI has a world lead.

These novel mapping technologies will also allow the creation of an expression map of the barley genome. We are in the process of characterising several thousand expressed sequence tags (ESTs) from the developing and malted barley grain. This gene discovery project aims to generate a more complete understanding of the patterns of gene expression which underlie the physiology and biochemistry of the developing endosperm and the barley grain during the malting process. Mapping of these sequences will allow their correlation to areas of the barley genome known, through QTL analysis, to contribute to characteristics such as malting quality. Ancillary data from tissue and temporal specificity, allelic diversity and its relation to the trait of interest, and information on metabolic pathways can all help to refine this candidate gene approach. Marker-assisted breeding or transgenesis will then be applied to test the function of the candidate sequence.

This approach relies heavily on the efficiency of the DNA sequencing facility which continues to provide an excellent service to researchers across the Institute despite an ever-increasing workload.

Cell biology Considerable progress has been made in the non-invasive imaging of virus movement in plants. Using viruses expressing GFP, the systemic pattern of invasion of potato virus X was studied in leaves undergoing the sink-source transition. Further studies of this developmental phenomenon have led to the discovery of two distinct populations of plasmodesmata, the channels through which viruses pass when infecting cells. Simple plasmodesmata, which occur only in sink tissues, permit the passage of macromolecules, while branched plasmodesmata, which form later in source leaf tissues, permit the passage of small solute molecules only. These developmental changes are correlated with the import/export transition of the leaf and demonstrate a major role for plasmodesmata in regulating assimilate fluxes in the plant (see pp. 76-79).

Collaborative research with Dr C. Hawes has been funded by the BBSRC and through the SOAEFD flexible fund, and is aimed at dissecting the secretory pathway in plant cells using virus-based vectors. This work has produced the first *in vivo* tags for the Golgi apparatus, an organelle involved in the sorting and redistribution of proteins within the cell, and is exploring the regulatory steps in the secretory pathway.

A collaborative venture between SCRI and Biosource Technologies Inc., a Californian-based biotechnology company, is exploring the use of viral vectors for the expression of foreign proteins in plants. This work involves a multidisciplinary approach utilising skills in molecular biology, virology, cell biology, and imaging. Eleven new appointments have been made to facilitate this major research programme.

A non-invasive study of the development of blackcurrant fruits from flower to maturity has been achieved by NMR microscopy, and the images compared with those derived from low temperature scanning electron microscopy (LTSEM) and resin histology. The entire living tissues of the specimen, still attached to the growing bush in some experiments, were imaged in three dimensions. By reference to *http:www.scri.sari.ac.uk* and clicking 'Special Topics', it is possible to see the animation of serial slices in selected planes and rotations of 3D projections to reveal internal structures of the flower and fruit, such as the ovaries, aril, vascular bundles and the mature seed. Because the NMR signal intensity is a function of mobile proton concentration and relaxation rate, it allows the generation of a variety of contrast patterns to reveal different aspects of structure without the use of stains. This work on the dynamics of fruit development is an example of the interdisciplinary nature of many advances in botany made possible by investment in new technologies at SCRI. The contrast patterns produced non-invasively by NMR imaging represent tissue features distinguished by entirely different physico-chemical processes from those revealed by light or electron microscopy and, at this stage in the development of NMR, it is important to make reference to illustrations produced by conventional histology.

The sink-source transition in leaves - new insights

A.G. Roberts, S. Santa Cruz, P. Boevink, I.M. Roberts, N. Sauer & K.J. Oparka

All leaves on a plant undergo a transition from a sink (a net carbon importer) to a source (a net carbon exporter) during their development. The early growth of a leaf is supported by carbohydrate imported from other sources in the plant. These sources are usually other mature leaves or photosynthetic organs on the plant, or in the case of a seedling, the cotyledons. As the lamina expands and the leaf matures, levels of photosynthesis increase until the leaf can support itself. When the amount of carbon accumulated by photosynthesis is greater than the requirement of respiration and growth, a positive carbon balance is achieved by that leaf. The leaf then becomes an exporter of carbon. In dicotyledonous plants, the conversion from sink to source begins shortly after the leaf begins to unfold, and is known as the sink-source transition. The transition begins at the leaf tip and moves basipetally as the leaf matures.

In *Nicotiana benthamiana*, five vein classes have been identified (Fig. 1). The class I vein (the midrib) branches at frequent intervals to form class II veins. These class IIs frequently interconnect at the leaf margins, providing a loop between adjacent class II veins. Class III veins are derived from class IIs and subdivide at regular intervals to form the class III veinal network. These three classes of veins are collectively termed the major veins, and previous autoradiographic evidence has shown that these are involved in unloading of photoassimilates from the phloem. Vein classes IV and V, the minor veins, are found within the islands of the class III vein network and are involved in phloem loading once the leaf has become a source.

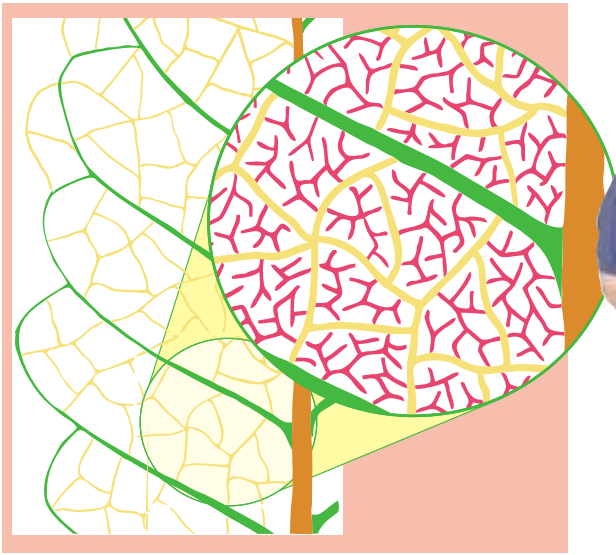

Figure 1 The veination of N. benthamiana. The class I vein (brown) branches to produce class II (green) and then class III veins (yellow). Minor veins (red) are found within the class III network.

Non invasive imaging of the sink-source transition

Until recently, the only way to image the sink-source transition was by using autoradiography. However, this method is destructive, technically demanding and time consuming. Recent studies carried out in the Unit of Cell Biology have shown that the sink-source transition can be studied more simply, and in real-time, with fluorescent tracers[1]. These techniques have enabled us to study the unloading pattern of photoassimilates using the phloem-mobile, fluorescent probe carboxyfluorescein (CF). This probe is loaded onto the abraded surface of source leaves as carboxyfluorescein diacetate (CFDA), which is then cleaved by plant esterases to produce the impermeant CF moiety. This is translocated through the phloem and unloaded in

sink leaves along with the flow of photoassimilate. After phloem import of CF, the leaves can be detached and their petioles placed into a solution of 3 kDa Texas Red dextran. This dye is transpired through the xylem network of the leaf and labels all veins, allowing the vein classes to be easily identified. The fluorescence in sink tissue is then detected and imaged using a confocal laser scanning microscope (CLSM).

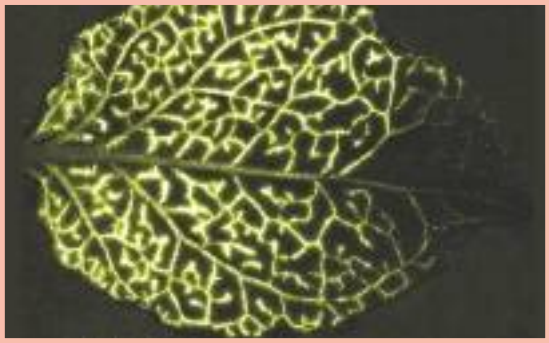

Figure 2 CF unloading from major veins in a leaf that has just started to undergo the sink-source transition.

The pattern of CF transport (Fig. 2) was dictated to a large extent by the phyllotaxy of the plant, with leaves directly above the loaded leaf becoming more uniformly labelled than leaves on the opposite side of the plant. In sink leaves displaying phloem transport of CF, dye was seen first in class I and II veins but was not observed to unload from these vein classes. In contrast, after entry into the class III veinal network, CF was unloaded into the mesophyll. In sink leaves in which the sink-source transition was absent, or was only recently commenced, dye unloading was widespread and the subtending tissues became highly fluorescent. However, in tissue close to the sink-source transition, CF unloading was greatly reduced and the dye was restricted to the class III veins. CF was not unloaded from the phloem in any source tissue or from class IV or V veins.

Virus movement follows the 'rules' of solute unloading Using the CF unloading studies as a basis, unloading of GFP-tagged viruses was then studied. Potato virus X (PVX) carrying the green fluorescent protein (GFP) was inoculated onto *Nicotiana benthamiana* leaves. The GFP allows the spread of the virus to be monitored in both whole plants and tissues when illuminated by UV light. PVX spread throughout the inoculated leaf and entered the phloem where it was carried in the flow of photoassimilate into sink tissues. The first indication of virus entry into sink leaves was the appearance of fluorescent flecks on the lamina, indicating that the virus was unloaded from discrete foci rather than uniformly along leaf veins (Fig. 3). Flecks of virus were first visible in sink leaves approximately 9 days after the plants were inoculated. After the appearance of fluorescent flecks, the mesophyll tissue between veins also became infected. Fluorescent images of five leaves from one systemically infected plant are shown in Figure 4. In sink leaves near the apex, the virus was present throughout the entire leaf, although the tips of these leaves often showed more intense fluorescence than did the base. By contrast, after the onset of the sink-source transition, the apical (source) region of the leaf showed no fluorescence, indicating an absence of virus. When Texas Red was introduced into the transpiration stream of systemically infected leaves, virus unloading was found to occur predominantly from the class III vein network; the same vein class used to unload solutes (Fig. 3).

Sink leaves can unload macromolecules We have been collaborating with N. Sauer's group in Erlangen, Germany. These researchers have expressed GFP in companion cells of source leaves using the promoter of the *Arabidopsis* sucrose transport protein, AtSUC2[2]. The GFP was produced in companion cells of source leaves, entered sieve elements, and was found to unload subsequently from the phloem in sink leaves. GFP was also found to spread from cell to cell in sink tissues. GFP is a 27 kDa protein and therefore much larger than the size exclusion limit (SEL) of plasmodesmata (see SCRI Ann. Rep. 1993,

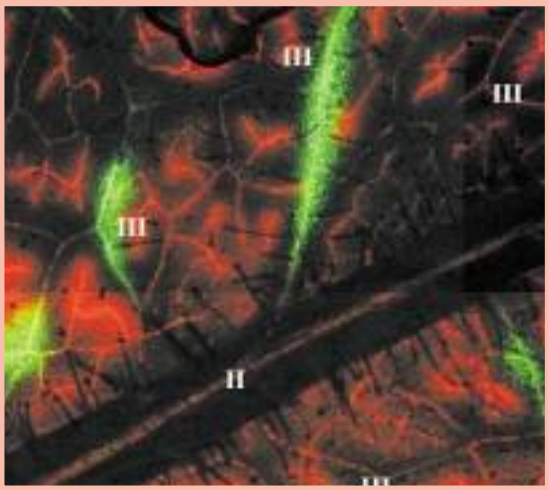

Figure 3 Unloading of GFP (green) occurs from the class III vein network and produces elongated flecks. The vascular system (shown in red) is labelled with Texas Red dextran.

Figure 4 Five apical leaves from a plant systemically infected with PVX.GFP. In small sink leaves, the tip of the leaf becomes more fluorescent than the base (arrowed). The largest two leaves were undergoing the sink-source transition at the time of virus entry.

50-53). Thus, some plant tissues have the capacity to traffick macromolecules.

The distribution of GFP allowed the sink-source transition to be studied non-invasively in intact leaves. GFP has a half-life in living cells of approximately 4 hours. This meant that, as the transition moved, alterations in the distribution of GFP between sink and source areas of leaves could be seen (Fig. 5). In source tissues, punctate fluorescence from individual companion cells could be seen along the veins due to the restriction of GFP to this cell type (Fig. 6). In contrast, sink leaves showed diffuse unloading of GFP from major veins (Fig. 7), characteristic of solute unloading (cf. Fig. 2).

Plasmodesmata in sink leaves have a SEL of approximately 50 kDa In order to probe the SEL of plasmodesmata in sink tissue, leaves were biolistically

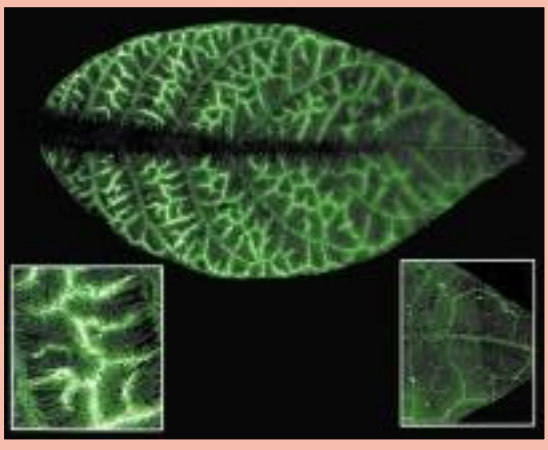

Figure 5 A leaf unloading GFP expressed from the AtSUC2 promoter. The sink source transition has just started at the leaf tip. Boxed areas from the tip and base of the leaf highlight the differences in GFP distribution in sink or source areas.

Figure 6 In source tissue, GFP is found only in companion cells, seen here in a section through a leaf. The xylem is autofluorescent.

bombarded with plasmids encoding various GFP-fusion proteins of different sizes (Fig. 8). Fusions were made between GFP and the storage proteins sporamin (mass 47 kDa), and patatin (mass 67 kDa). A third fusion was created between GFP and a truncated version of the patatin protein (GFP-patati), which had a mass of 61 kDa. When leaves were bombarded with a plasmid expressing only the *gfp* gene, intense fluorescence was detected in the initial bombarded epidermal cell and also in several cells surrounding the 'hit' cell. GFP was detected in neighbouring cells within 7h, and 2 days later had spread into approximately 200 cells (Fig. 8A), moving upwards into leaf trichomes and also downwards into the mesophyll. In contrast, when GFP was bombarded into epidermal cells of source leaves, the spread of fluorescence was greatly restricted (Fig. 8B). However, trace movement was often observed in cells immediately adjoining the bombarded cell. In sink leaves, the GFP-sporamin fusion moved several cells away from the bombarded cells, but not as extensively as GFP alone. In contrast, no cell-to-cell movement of GFP-sporamin was

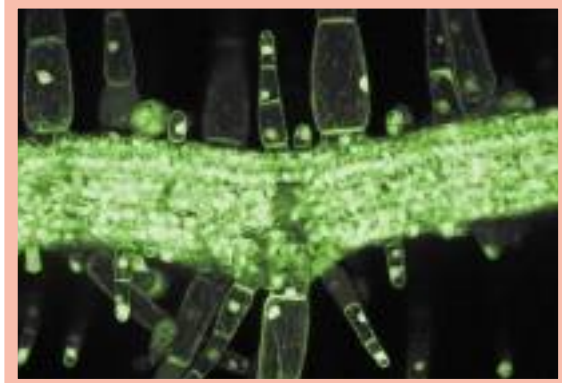

Figure 7 In sink tissue, unloaded GFP is present throughout the cytoplasm of each cell in the leaf.

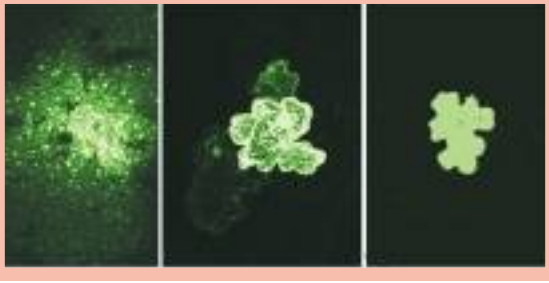

Figure 8 Leaf tissue bombarded with GFP and GFP-fusion proteins. (A) In sink tissue, GFP moves rapidly from cell to cell. (B) In source tissue, GFP is much more restricted to the bombarded cell. (C) In sink tissue, GFP-patatin does not move out of the bombarded cell.

observed in source leaves. Both the GFP-patatin (Fig. 8C) and the GFP-patati fusion proteins failed to move from cell to cell in either sink or source leaf tissues. These results demonstrate that proteins of at least 47 kDa, but not as large as 61 kDa, can move freely through sink-leaf plasmodesmata. This plasmodesmatal permeability decreased in response to the developmental switch from sink to source status.

The sink-source transition is accompanied by a change from simple to branched plasmodesmata We have shown that, during the sink-source transition in tobacco leaves, simple plasmodesmata give rise to more branched forms as the leaf matures. This provided an ideal system to study the functional differences between simple and branched plasmodesmata. To examine the structure of plasmodesmata during the sink-source transition, leaf samples were taken from each leaf on AtSUC2 transgenic tobacco plants expressing GFP. Before excision of the samples, the position of the sink-source transition was recorded by imaging each intact leaf under the CLSM using low magnification objectives. The sink-source transition was demarcated by the change in appearance of vein classes that were unloading GFP and those that showed CC-specific expression of GFP (see Fig. 5). Counts of plasmodesmal types (simple versus branched) revealed a marked reduction in the proportion of simple plasmodesmata in source tissues during the progression of the sink-source transition (Fig. 9). Tissue samples taken at the base of the leaf showed predominantly simple plasmodesmata, while those at the tip were mainly branched. In samples studied by electron microscopy, the simple plasmodesmata had conspicuous wall collars surrounding the neck of the pore. Within the region of the sink-source transition, 'pairs' of simple plasmodesmata were observed routinely in both longitudinal and oblique sections of the

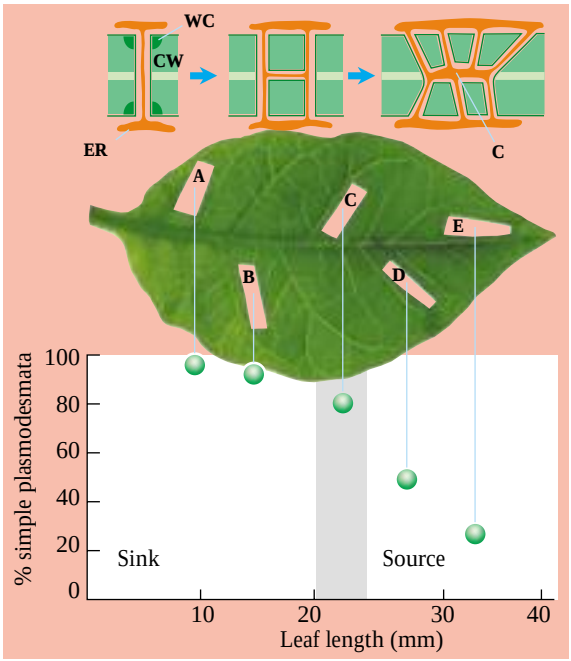

Figure 9 The sink-source transition is accompanied by a change from simple to branched plasmodesmata (top). The graph (bottom) shows the percentage of simple plasmodesmata in different sectors of the leaf (A-E). The transition zone is shown within the shaded region. C, central cavity; CW, cell wall; ER, endoplasmic reticulum; WC, wall collar.

wall, giving rise to 'H'-shaped plasmodesmal branching patterns. Branched plasmodesmata also displayed conspicuous median cavities. Unlike the simple plasmodesmata in sink tissues, the branched plasmodesmata found in source regions of the leaf did not display electron-dense wall collars.

Our studies have shown that the sink-source transition is a dynamic change that occurs in all leaves and that it has an enormous effect on both the structure and function of leaf tissues. Sink leaves are capable of non-specific trafficking of molecules of at least 47 kDa through simple plasmodesmata. On the other hand, the branched plasmodesmata in source tissues have a low size exclusion limit and restrict macromolecular trafficking. This suggests that the leaf utilises a 'downregulation' of plasmodesmal conductance to restrict intercellular communication during the sink-source transition. It thus appears that the plant uses plasmodesmata as 'control centres' for macromolecular trafficking.

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Plant genes for the spliceosomal protein, PRP8

J.I. Hamilton, C.G. Simpson, G.P. Clark, C. McQuade, J.M. Lyon, J.A. Watters & J.W.S. Brown

The majority of plant genes contain intervening sequences (introns) which interrupt the protein-coding sequence. Following transcription into precursor messenger RNA (pre-mRNA), the introns must be removed to generate the mature mRNA, which is then translated into protein in the cytoplasm. Removal of introns is known as splicing and occurs in a large complex of RNA and proteins called the spliceosome. The spliceosome contains in the region of 100 proteins. Many of these proteins are evolutionarily conserved and are known to be essential for the splicing process in yeast and animal systems. Nevertheless, even in such highly conserved biochemical machinery, there are differences between plants and other eukaryotes in terms of components. One such component, which is known from yeast to be essential for splicing, is the spliceosomal protein, PRP8.

One of the exciting areas to emerge in the splicing field over the last 3-4 years has been the discovery of a class of introns which differ from conventional introns. The former, known as AT-AC introns, are removed by a different spliceosome (Fig. 1). The major spliceosome, which removes conventional introns, contains four main components, termed small nuclear ribonucleoprotein particles or snRNPs: U1, U2, U4/U6 and U5. The minor spliceosome, which removes AT-AC introns, contains U11, U12, $U4_{atac}/U6_{atac}$ and U5snRNPs. As a result, the spliceosomes are called U2-dependent spliceosomes and U12-dependent spliceosomes respectively. While many components differ between the two spliceosomes, it is noteworthy that the U5snRNP is utilised in both, and that PRP8 is a U5snRNP protein.

	5' splice site	Branch point	3' splice site
U2	GU	CURAY	AG
U12	AU GU	UCCUUAAC UCCUUAAC	YYCAG YYCAG

Figure 1 Conserved sequences in U2-dependent and U12-dependent introns.

PRP8 was discovered first in yeast, where at 280 kDa, it is one of the largest proteins in the yeast cell. PRP8 has also been isolated from man, *Caenorhabiditis elegans*, *Arabidopsis* and maize. Maize *PRP8* genes were isolated at SCRI on the basis of conserved sequence. The *Arabidopsis PRP8* gene was isolated by colleagues in Oklahoma State University, USA. These represent the only plant *PRP8* genes cloned to date and we have used them to investigate gene number, expression and potential function of the genes.

Genomic and cDNA *PRP8* clones have been isolated from maize (Fig. 2). The maize gene contains 13 exons and 12 introns, spanning 9.8 kb of genomic sequence. The gene would encode a protein of 2,363 amino acids. The *Arabidopsis* PRP8 gene was isolated from a T-DNA insertion mutant, *sus2-1*, which gave an embryo-lethal phenotype. The isolated gene was able to complement the *sus2-1* mutant, showing that it was functionally equivalent. The coding regions of these two plant genes were over 98% identical at the amino acid level. However, they differed in two significant ways. First, the maize N-terminal region was proline-rich, as found in yeast PRP8, while *Arabidopsis* and *C. elegans* PRP8 proteins did not contain this region (Fig. 2). The proline-rich N-terminal region of yeast is essential for splicing, making the difference between the plant PRP8 proteins intriguing. Sec-

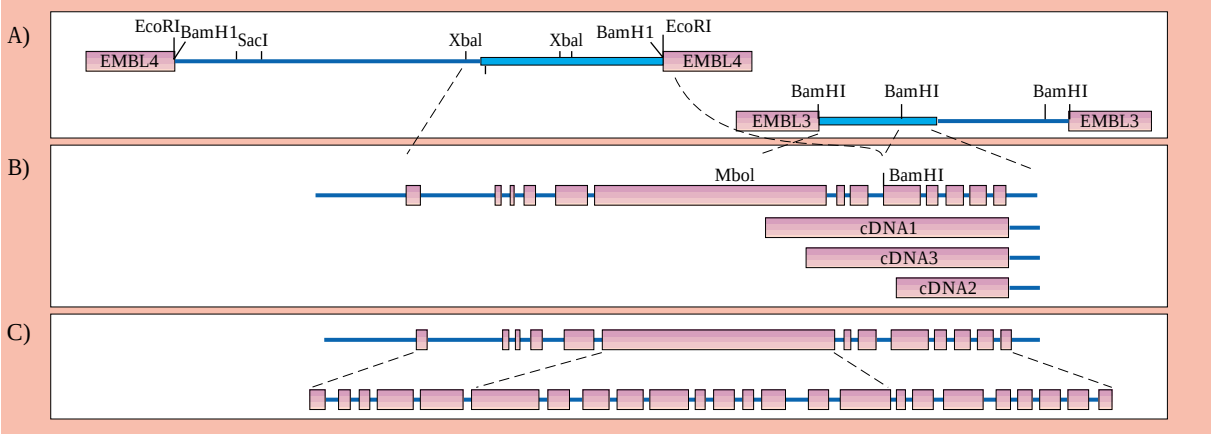

Figure 2 Comparison of gnomic structure of *PRP8* genes isolated from maize and Arabidopsis. Boxes represent exon (coding) sequences which are interspersed by introns (lines)

ond, the *Arabidopsis PRP8* gene contained 23 introns, 11 more than that of maize. Ten of the extra introns in *Arabidopsis* interrupted a region which in maize *PRP8* constituted the large central exon. This finding is contrary to the widely-held view that *Arabidopsis* genes are less complex than those of other plants.

Gene number was analysed by Southern analysis of maize and *Arabidopsis* genomic DNA and by screening *Arabidopsis* yeast artificial chromosome (YAC) libraries. In both, *PRP8* genes are present in small multigene families of 3-5 copies and those in *Arabidopsis* were located on chromosomes 1 and 4. Thus, plants differ from human and yeast systems, which contain only a single *PRP8* gene. That there are 3-5 *PRP8* genes in *Arabidopsis*, but mutation of a single gene in *sus2-1* leads to embryo death, raises further questions on the expression patterns of the different genes or functional diversity. Clearly, in human, the same PRP8 protein will be present in both U2-dependent and U12-dependent spliceosomes. However, the presence of multiple genes in plants, the observed differences between the cloned *Arabidopsis* and maize genes and the phenotype of *sus2-1*, raise the possibility that different PRP8 proteins are found in the U2- and U12-dependent spliceosomes in plants.

At SCRI, we have obtained two lines of evidence to suggest that is the case. First, at least two different *PRP8* genes are expressed in developing embryos at the time when the *sus2-1* embryos abort. Thus, PRP8 protein is likely to be present. Second, in *sus2-1* mutant embryos, splicing of conventional introns is unaffected, suggesting that the *prp8* mutation in *sus2-1* has not affected U2-dependent splicing. Our current hypothesis is that plants contain multiple *PRP8* genes, which encode proteins functioning in either U2- or U12-dependent spliceosomes. The genes are different in exon-intron structure and are assembled into the different spliceosomes on the basis of the presence or absence of the polyproline N-terminal

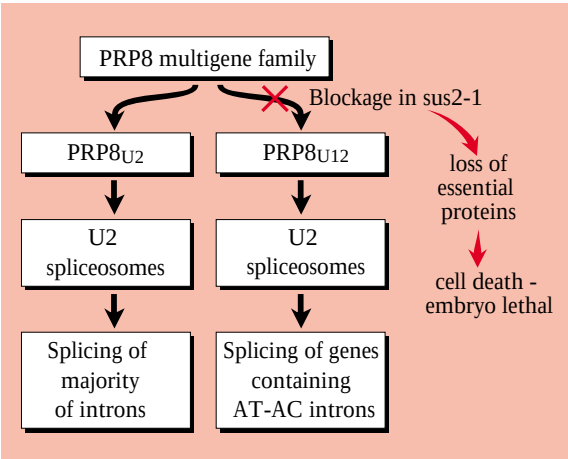

Figure 3 Model for embryo-lethal phenotype of *Arabidopsis sus2-1* mutant.

region. In *sus2-1*, the mutant *prp8* gene may encode a U12-dependent PRP8. If there is only one such gene (reflecting the much lower abundance of U12-dependent spliceosomes), the mutation will knock-out U12-specific PRP8, leading to lack of splicing of all AT-AC intron-containing transcripts. This would lead to embryo death, even in the presence of U2-specific PRP8, U2-dependent spliceosomes, and unaffected splicing of normal introns (Fig. 3). We are currently investigating splicing of AT-AC introns in *sus2-1* mutant embryos and are investigating further the characteristics of individual gene members in the maize and *Arabidopsis* families.

Promoting plant promoters

G.C. Machray, P.E. Hedley, D. Davidson, A.F.M. Ibrahim, J.A. Watters & J.W.S. Brown

Agricultural application of plant genetic manipulation is now a reality, with major shares of production of a variety of crops in the USA being given over to new cultivars produced through recombinant DNA technology. The technology is still in its infancy however, being confined largely to the modification of single traits, involving the transformation of plants with single genes. Many of these confer entirely new properties to the plant, additional to and largely independent of the existing plant biochemistry and physiology. Others, while similar to existing functions, have properties which confer insensitivity to the regulatory checks and balances to which their endogenous plant analogue is subject, and hence superimpose the desired effect on a background of normal plant cell metabolism. In the main, reactions at the end of complex pathways have been targeted for manipulation to yield a desired product. Such approaches, while successful, offer limited insight into the grander scheme of plant metabolism, which remains an area of great opportunity for more sophisticated applications of this technology, both for the increased knowledge of plant biology to be gained, and for the subsequent rational and safe exploitation of that knowledge.

Many factors need to be taken into account to refine and improve strategies for the genetic manipulation of plant cell metabolism. Pathway control may be shared among a number of components and require treatment as a quantitative trait. Intricate regulatory networks, mediated by multiple signal transduction pathways, will require dissection and individual manipulation of contributing elements. The effects of the compartmentation of metabolism and metabolic channelling must be considered. All of these require detailed knowledge of relevant gene expression and of the fate of the products of that expression. This information will aid decision-making in the choice of targets for manipulation. In addition to the identification of targets, effective manipulation of the complex metabolic processes which are targeted will require highly specific promoters able to regulate gene expression in a highly controlled manner, to ensure that spatial and temporal constraints are met.

Promoters can be divided into two major classes: constitutive, which are expressed in all cells, and regulated, which can be expressed in particular cells or tissues, or at particular stages of development. In addition, expression levels from each type of promoter can be highly regulated. The source of promoters for use in transgenics is isolated genes. Many plant genes are organised in multigene families where variation in

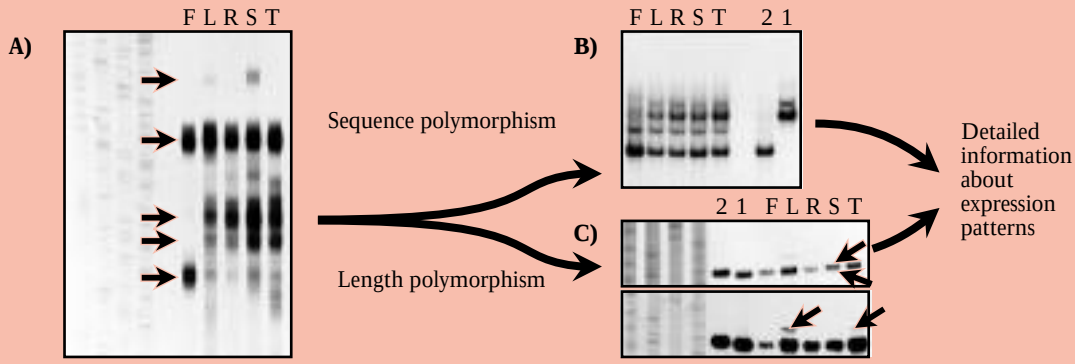

Figure 1 A novel approach for expression profiling of multigene families and isolation of gene-specific promoters
5' RACE is carried out on total RNA isolated from different organs using a [^{32}P] end-labelled primer. Products are separated on a DNA sequencing gel alongside a DNA sequencing ladder marker (A). Products are either directly cloned into pGEM-T Easy and sequenced, or individual bands (arrows) are excised, re-PCR'd and sequenced. The generated sequences of the 5' UTRs allow specific bands to be related to different genes and/or different transcription start sites. If there is sequence polymorphism but no length polymorphism among different gene family members (U1A), SSCP analysis is carried out by running RT-PCR products generated by using end-labelled primers on a mutation detection enhancement (MDE) gel (B). Labelled PCR products from plasmids representing individual members are used as controls (1=U1A-1, 2= U1A-2). If length polymorphism results from different sequences of the 5' UTRs (U2B"), labelled RT-PCR products are separated on a DNA sequencing gel (C) with labelled PCR products from clones of individual members as controls (1=U2B"-1, 2=U2B"-2). Short exposure allows visualisation of single base differences (arrows in upper photograph), while longer exposure and higher loading intensity allow detection of rare transcripts (arrows in lower photograph). The detailed information from these experiments allows primers to be designed for the isolation of gene-specific promoters by inverse PCR. F=flower, L=leaf, R=root, S=stem, T=tuber.

expression pattern and level exists among individual members. We have been actively developing and applying molecular techniques to examine expression pattern and rapidly isolate gene-specific promoter sequences.

Differential spatial and temporal expression from complex gene families can be dissected using technologies such as RT-PCR[1] (reverse transcription - polymerase chain reaction) based on differences in length of transcripts. Where no length variation exists, SSCP (single-stranded conformational polymorphism) based on variation in coding sequence content, can be applied[2]. Many genes also encode transcripts which include non-coding regions - untranslated regions at their 5' or 3' ends (5'-UTR, 3'-UTR). The nucleotide sequences of these can be obtained by approaches such as RACE (rapid amplification of cDNA ends). These sequences are ideal for RT-PCR techniques, because they are more variable, both in length and content, than the coding sequence for genes which are members of gene families. Use of gene-specific sequences for individual gene family members in these approaches can deliver not only increased definition of the tissue-specific expression patterns from multigene families but can also confirm constitutive expression. 5'-UTR sequences, as well as 5' introns, can be used in the design of primers for inverse-PCR (IPCR) from the genome to obtain specific promoter sequences for genes which show constitutive or regulated expression. These promoters then provide the switches to deliver constitutive expression, or to manipulate expression specifically within selected tissues, of heterologous genes when built into recombinant constructs.

Within the Gene Expression Unit, these RT-PCR based methods have been used to characterise expression from several complex plant gene families and to clone promoters from genes which show interesting expression profiles. Figure 1 describes application of these technologies to the analysis of tissue-specific expression of the U1A and U2B" gene families in potato. This has revealed lack of expression of one U1A gene in floral tissue, constitutive expression of another, and constitutive expression of an additional gene not previously detected. Two U2B" genes were shown to be constitutively expressed, while a third was expressed only in leaf and tuber tissue. The confirmation of constitutive expression of four genes is in itself

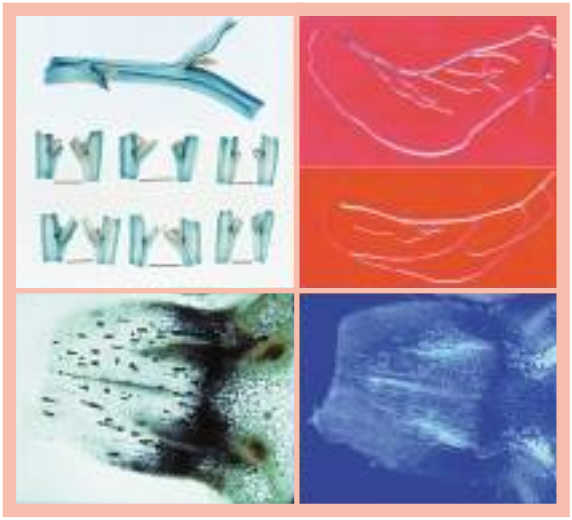

Figure 2 Expression pattern from the promoter of the invertase gene *invGE*. GUS reporter gene expression driven by the *invGE* promoter in a transgenic potato line under axillary buds in stem (top left) and in transgenic v. control roots (top right). The paired images below show the same cross section of a stem axil from this transgenic line stained with GUS (left) and with aniline blue (right) to reveal callose-containing xylem tissue.

an important finding - the promoters of these genes are candidates for genetic manipulations requiring constitutive expression. One which we have cloned is expressed at 65% of the level of the CaMV 35S promoter; promoters such as this are of increasing importance given concerns over gene silencing resulting from promoter duplication and the use of promoters derived from viruses or other plant pathogens.

These methods also have been applied to analyse tissue-specific expression and to obtain promoters from the invertase gene family in potato. We have previously described the characterisation of a pollen-specific invertase promoter[3] and suggested a potential use for the pollen-specific promoter in the generation of male-sterility in cultivated potato. This is now perhaps an even more desirable trait given current concerns over pollen dispersal into the environment on release into field-trialling of genetically-manipulated crops. Immediately upstream of this promoter in the potato genome lies a second invertase gene. The promoter of this second gene has been cloned by the methods described above and a series of transgenic potato plants generated in which it drives expression of the *uidA* gene coding β-glucuronidase (GUS). This promoter also determines expression in floral tissues, including pollen and the calyx. Its expression pattern is more diverse however and further expression is seen in specific vegetative tissues of the potato plant. Notably, these include regions where the vasculature branches, such as where the stem gives rise to lateral leaves or stolons, or where lateral roots branch from a main root (Fig. 2). Expression under the lateral bud of the stem is important because in the tuber, which is a modified stem with shortened and broadened axis,

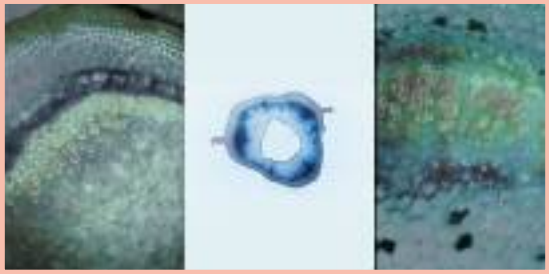

Figure 3 Expression pattern from the promoters of the invertase genes *invCD1* and *invCD4*. Both promoters drive expression in the three major and three minor vascular bundles and associated tissues, as seen in the GUS-stained cross-section (center) of stem tissue from a transgenic potato line carrying an *invCD1* promoter-GUS reporter fusion. Individual vascular bundles from the appropriate transgenic lines co-stained for GUS activity (blue) and with aniline blue to reveal callose-containing xylem (white) showed *invCD1* expression preferentially in external phloem (left) and *invCD4* expression preferentially in internal phloem (right).

the equivalent of this region is the 'eye', which will give rise to the new sprout. Each eye is formed by leaf scars with subtended lateral buds with undeveloped internodes, and expression was also detected from this promoter in eyes excised from tubers. The upstream promoter may have utility in the control of developmental processes in the potato plant since it governs expression in key tissues for micropropagation as well as in the sprouting of tubers. Enhanced control of these processes could have significant commercial benefits.

Two further invertase gene promoters have been cloned using gene-specific IPCR technology. Series of transgenic lines generated for each of these, in which they govern expression of *uidA*, indicate a highly specific expression pattern confined to stem and petiole tissues. Both promoters direct expression in vascular tissue, within the three major and three minor vascular bundles and further vascular tissue interspersed between them (Fig. 3). In potato, each vascular bundle is composed of external and internal phloem sandwiching a layer of xylem. When examined in detail, one promoter expressed GUS preferentially in external phloem while GUS expression was driven preferentially in internal phloem by the second promoter. Both promoters may have application in insect control strategies, particularly against sap-sucking insects. Further roles for these promoters are under investigation - evidence suggests they are, for example, likely to be switched on by pathogen attack and may therefore have further use in control strategies targeted against diverse plant pathogens ranging from nematodes to fungi and bacteria.

We have described the successful application of these methods to the analysis of differential expression in diverse plant multigene families and to provide promoter switches for further biotechnological application. While this is an important objective, there also remain interesting biological questions about the promoters and the genes which they regulate. What are the controlling elements in the promoters and can they be manipulated to generate altered expression profiles? What are the roles of the gene products in the biochemistry and physiology of the plant and why have multiple genes evolved to carry out this function in closely-related environments? Fortunately, the promoters themselves provide a means by which we can begin to address these problems by driving targeted antisense or post-transcriptional gene silencing knock-out of the function of their cognate gene.

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Transparent plants: an NMR case-study of blackcurrants

S.M. Glidewell, B. Williamson & G.H. Duncan

Imagine the advantages of being able to view a living, growing plant in 3D, with the freedom to select contrast to reflect different physico-chemical states of tissues within the specimen. NMR microimaging provides a non-invasive method to look at the internal structure of plants and therefore to study living tissues as they function, change, grow, age, or become affected by stress or diseases.

Until recently, the changes taking place during internal plant development could be studied only by histological methods based on approaches devised last century. Destructive sampling of a population of individuals was followed by chemical fixation of specimens to stabilise proteins, desiccation in fluids and embedding in wax or resin. With the rigidity provided by the infiltrated wax or resin, the specimen could then be sectioned thinly and stained to add contrast before examination by transmitted light in conventional microscopy. Despite important advances in microtomy and microscopy (see Ann. Rep. 1994, 172), only relatively small specimens can be handled by most of these methods. Unfortunately, at the end of tissue processing, the specimen has changed its dimensions markedly and many important plant constituents (e.g. oils, waxes, gums and resins) have been extracted unintentionally. If a 3D depiction of the internal structures is required, the sections must then be photographed and the images reconfigured to the original shape.

NMR imaging provides a powerful non-invasive means to look at the internal structure of plants. In addition, the technique circumvents problems encountered with attempting to section extremely tough specimens (e.g. woody stems, nuts, and ripe fruits containing hard seeds). Since no light is involved in the imaging process, there is no size limitation on the samples, other than the bore of the super-conducting magnet. Although NMR imaging is recognised as a powerful technique which reveals many anatomical features unambiguously, at this stage in its development for uses in botany, the identity of many tissue features needs to be corroborated and the origin of the contrast in the image understood.

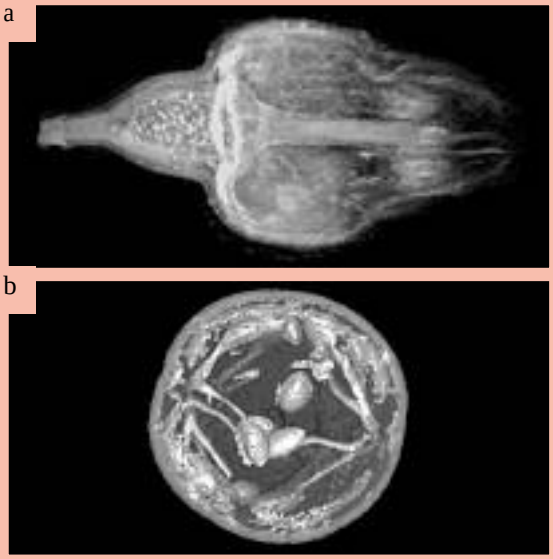

Figure 1 a) Maximum intensity projection of a closed blackcurrant flower b) Surface rendered image of an entire ripe blackcurrant fruit – 'electronically cut' to reveal interior detail

For instance, a recent study[1] of blackcurrant fruits from flower (*Fig. 1a*) to maturity (*Fig.1b*) revealed very clearly the vascular bundles in the periphery of the flower (*Fig. 2a*) and the ovary (*Fig. 2b*). These bundles were equally clear in the mature fruits (*Fig. 2c*). However, the relative intensities have reversed. This is not an artefact, as images acquired with a wide range of parameters (which can lead to contrast reversals depending on relative relaxation times) consistently produced images of flowers where the vascular bundles had high (bright) intensity in a dark field and mature fruits where the reverse was true. Only with the

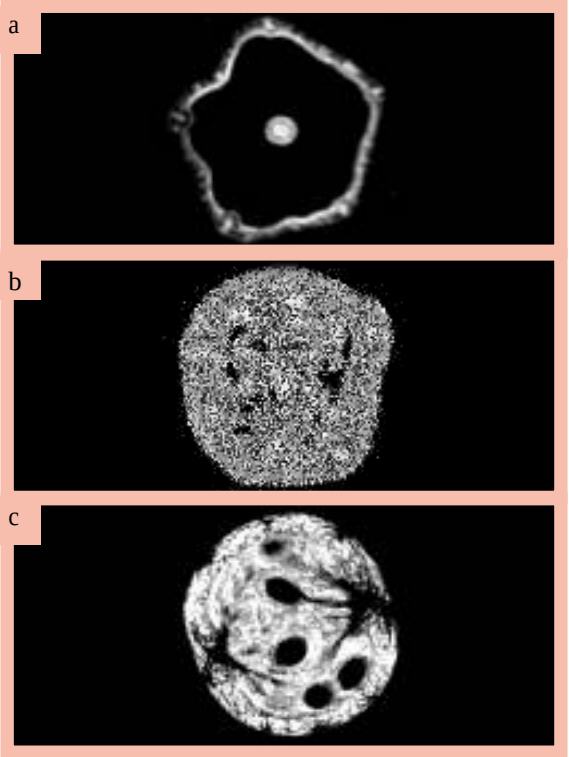

Figure 2 a) Transverse plane of closed blackcurrant flower b) Median transverse plane of ovary of blackcurrant flower c) Transverse plane of ripe blackcurrant fruit.

use of other microscopical techniques in tandem with the NMR was the apparent contradiction resolved.

SEM pictures of freeze-fractured ovaries (*Fig. 3a)* showed that the dark areas around the ovules in the NMR images (*Figs. 3b, 2b)* were indeed empty, whereas resin-embedded sections viewed by light

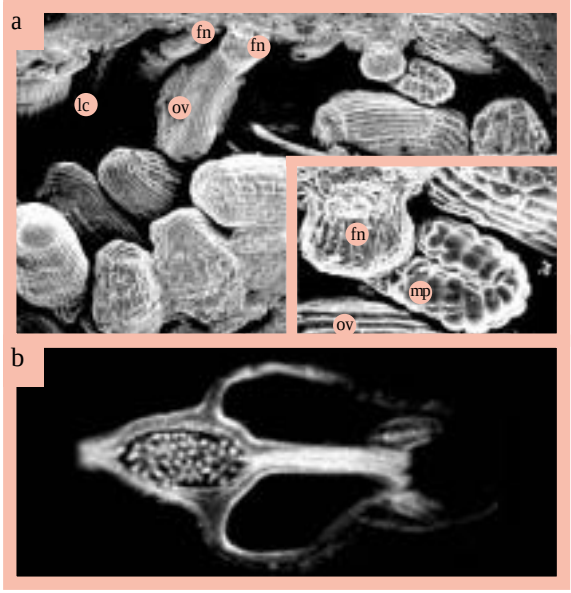

Figure 3 a) LTSEM of ovary of blackcurrant flower. Inset – close up of micropylar region. *lc – locular cavity, fn – funiculus, mp - micropyle, ov – ovule* b) NMR median longitudinal plane of closed blackcurrant flower.

microscopy of green fruits showed that the black vascular bundles in the NMR images corresponded to parenchyma cells separated by air spaces around a small vascular bundle which corresponded in size with the bright core at the centre of the NMR images of the 'bundle'. The phenomenon of

Nuclear Magnetic Resonance (NMR) imaging works by placing the sample in a strong magnetic field (inside a large magnet (shown in the picture)). This causes a tiny proportion of certain atomic nuclei to move to a higher energy level. A pulse of radiofrequency energy upsets this equilibrium and causes the nuclei to precess in phase. After the pulse is over, the system relaxes back to equilibrium, emitting radiofrequency energy. The amplitude of this emitted energy and time at which it is detected depend on the number of nuclei present and the rate at which they relax.

Most imaging experiments use hydrogen nuclei (protons) and the commonest proton-containing molecule is water. So NMR is imaging mostly water, but also other mobile protons such as those in lipids. In addition to the mobile proton distribution in a plant tissue, the signal intensity depends very strongly on the relative relaxation times of the protons which in turn are determined by the environment in which they find themselves. This can, for example, be water closely bound to large molecules such as polysaccharides or proteins, water in cell vacuoles which contain dilute solutions of many species, water in viscous gums or lipids in seed endosperms. In addition, factors such as diffusion, compartment size and tissue inhomogeneities can also affect the relaxation rates.

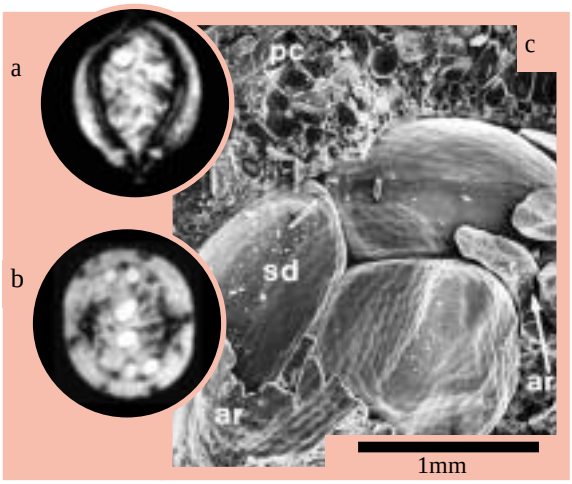

Figure 4 These two NMR images (a, b) are planes of the same small green fruit attached to a bush. As well as showing the aril network, the seeds can be seen as bright images and the bright vascular traces supplying the placentae are clearly evident in the longitudinal plane (a). c) LTSEM of small green fruit. *pc – pericarp, sd - seed, ar – aril.*

reduced intensity in NMR images due to interfaces between gas spaces and tissues is well understood. The interpretation of these botanical specimens therefore is confirmed and the NMR images validated for further work

The pulpy tissues around the maturing seeds constitute the aril, finger-like parenchyma tissues growing

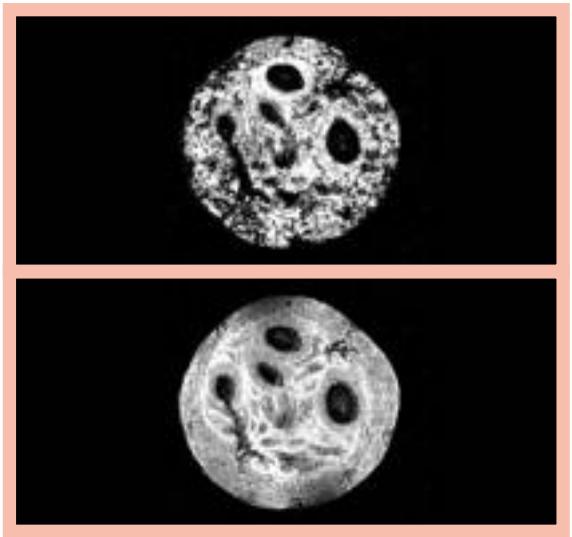

Figure 5 NMR images, using different protocols, of a large green fruit. By this stage, the seeds have hardened and appear dark. The gelatinous sheaths can be seen around them and the funiculi connecting them to the placentae which, along with the peripheral vascular bundles, are still bright.

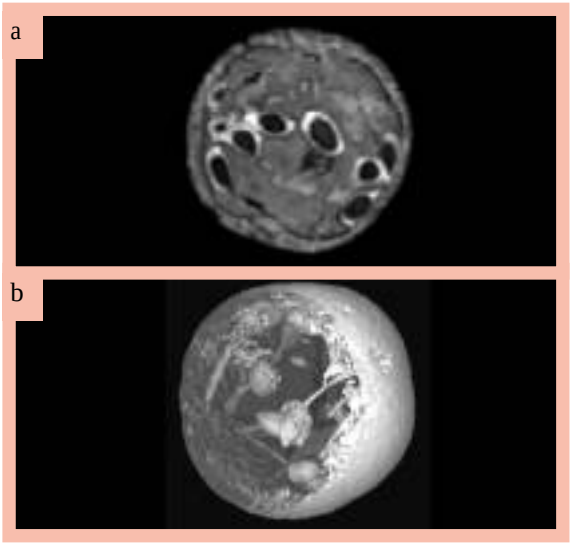

Figure 6 a) Median transverse plane of a ripe blackcurrant fruit. b) Surface rendered image of ripe fruit.

from the placentae to fill the spaces between seeds in the locular cavity. In NMR images (*Figs. 4a,b)*, the aril was displayed as a 3D network of bright strands, an interpretation confirmed by LTSEM (*Fig. 4c).*

Seed maturation involves the growth of a thick hard seed coat, deposition of lipid reserves in the endosperm and retention of a gelatinous sheath around each seed. All these features were resolved and studied non-invasively by NMR imaging (*Fig. 5)* and confirmed by conventional histological methods. By the time full ripeness is achieved, the fruit is too squashy and the seeds too hard for conventional sectioning. NMR images show very marked contrast between the dark seeds and bright gelatinous sheaths surrounding them (*Fig. 6a)* to such an extent that a surface-rendered 3D image of the inside of the fruit can be derived (*Fig. 6b*).

To appreciate the full 3D effect of these images, visit our website at *http://www.scri.sari.ac.uk* and click on 'Special Topics' where you can journey through a blackcurrant fruit or see right inside a closed flower or ripe fruit as it rotates.

Acknowledgements

Part of this work was done in collaboration with J. A. Chudek and G. Hunter of the Department of Chemistry, University of Dundee.

At SCRI, the work was funded by SOAEFD and the NMR imager was purchased by Mylnefield Research Services, Ltd.

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Breeding and genetics

George R. Mackay

Genetic variation or biodiversity within crop species is an essential pre-requisite for continued production of new improved cultivars. This is particularly so if those new cultivars are to be more resistant to pests and pathogens and less reliant on routine prophylactic agrochemicals, to maintain their yield and quality. The European form of the potato, Solanum tuberosum ssp. tuberosum, represents perhaps a classic example of a crop species which lacks genetic variation and, consequently, requires substantial inputs of agrochemicals to maintain its productivity. The sustainability of this approach is questionable for a crop which ranks fourth in world importance after wheat, maize and rice, in terms of human food production.

The potato is a relatively recent introduction into Europe, having been introduced during the Spanish conquest of South America about 500 years ago. These early introductions are believed to have been of the subspecies *andigena*, still cultivated in its native Peru. Adapted to the short days of equatorial regions, it took approximately 200 years for *andigena* to evolve into the long day adapted *tuberosum* form which predominates world-wide today. Limited introductions of *andigena*, and the loss of genetic variation that will have taken place during its evolution in Europe, meant that *tuberosum* offered no resistance when late blight was introduced from Mexico to Europe in the mid-19th century, and caused the catastrophe that halved the population of Ireland. More recently, as potato cyst nematode populations increased due to shortened rotations in the mid-part of this century, it was quickly found that no European forms of potato possessed resistance to this serious pest.

Recognising *tuberosum's* narrow genetic base, breeders have successfully introgressed genetic resistance from several of the many related wild species and primitive forms that exist in Central and South America. Perhaps the most striking example was the rapid introgression of the H_1 gene, conferring resistance to the golden cyst nematode (PCN) pathotype RO_1, from ssp. *andigena* CPC1673, into *tuberosum* and now available in most modern cultivars. Unfortunately, introgression of more complex genetic resistance to

the white cyst nematode, from more distantly related wild species such as *Solanum vernei*, has taken much longer and is only partially effective.

SCRI researchers are fortunate to have immediate access to the Commonwealth Potato Collection (CPC) of approximately 1,400 accessions representing more than 80 species of the tuber-forming members of the genus *Solanum*. The conservation and maintenance of such *ex situ* gene banks is particularly important when access to the wealth of genetic diversity in Central and South America is time consuming and costly due to the need to passage any *Solanum* material from the Americas through quarantine. However, conservation and maintenance is of little value if the germplasm is not properly characterised and utilised. At SCRI, though good levels of resistance to late blight and potato cyst nematodes are being achieved in recently released cultivars, and combined effectively in the multi-trait breeding experiment, we are conscious that our sources of resistance are limited to rather few species. It is strategically very important that additional, hopefully novel, sources of resistance are found and introgressed into *tuberosum*, lest the currently available sources are defeated by new strains of pest or pathogen.

Recently obtained external funding from the EU and an SOAEFD Flexible Fund project have provided the opportunity to expand the search for new sources of resistance to both late blight and PCN in the CPC. To date, sources of resistance to late blight have been identified in 57 accessions belonging to species from the seven taxonomic series within the tuber-bearing part of the genus *Solanum*. Not surprisingly, many of these are from Mexico, centre of origin of late blight itself, but several are from Bolivia and Argentina.

Potentially, *Solanum papita*, a tetraploid Mexican wild species, seems to be a very useful source and has been successfully hybridised using mentor pollen and embryo rescue with a tuberosum cultivar. It was intended that, with molecular marker aided selection and rapid screening for resistance of the backcrosses to tuberosum, this complex form of resistance could be introgressed rapidly into agronomically-adapted tuberosum. However, the resistance tests on the F_1 and backcross populations produced so far are proving difficult to interpret. Further crosses between resistant and susceptible accessions of *papita* itself are planned, so that the genetics of this potentially novel form of resistance can be interpreted. In the meantime, however, a diploid population of the species *S. verocossum* produced from crosses between susceptible and resistant accessions, is proving most useful. Being diploid and self-compatible makes this species ideal for genetical research and several molecular markers linked to QTL for late blight resistance have already been identified. This material will also prove ideal to investigate the feasibility and efficacy of molecular marker-aided selection at the diploid level. Similarly, at the tetraploid level, molecular markers linked to a major QTL for PCN resistance derived from *S. vernei*, have been located on linkage group IV. This QTL is only present in cultivars containing *vernei*-derived resistance and will also be extremely useful in establishing the effectiveness of marker-aided selection for *vernei* resistance at the tetraploid level.

Potentially useful, high levels of resistance to the bacterial *Erwinias*, which cause soft rot in tuber have been identified in the cultivated diploid group *phureja*. As these are clones from the unique SCRI long day adapted population of *phureja*, introgression into *tuberosum* should be relatively easy and the diploid status of *phureja* also facilitates studies into the genetics of resistance. Some clones from crosses between two soft rot resistant parents are also showing good levels of resistance to blackleg.

In addition to proving a valuable source of resistance and a strategically important resource for fundamental research into genetics, SCRI long day phurejas may have a direct commercial potential as flavoursome, high value novelty potatoes. In 1998, two *phureja* clones were submitted as potential cultivars for National List Trials on behalf of two private sector organisations, who have been funding the routine testing, trialling and selection through MRS Ltd.

Classical hybridisation and backcrossing to introgress genetic variation from wild species of *Solanum* into *tuberosum* can be time-consuming or not possible, particularly if the species have a different endosperm balance number (EBN) to *tuberosum*. Differing levels of ploidy and other factors can also result in sterility

problems, which hinder or prevent progress. Bridging crosses, embryo rescue and artificial manipulation of ploidy (with colchicine) can overcome some of these difficulties. The use of marker-aided selection may speed up the process by reducing the number of back-cross generations, but is yet to be proven. An alternative is to circumvent all the problems of sexual hybridisation by bypassing it altogether. We have been very successful at developing somatic fusion as an asexual means of hybridising wild species with *tuberosum*. In 1998, 283 protoplast-derived clones were grown in small, 4-plant plots at Blythbank. Fifteen per cent were selected for retrialling in 1999 on the basis of their agronomic potential, compared with control cultivars. These selected clones included somatic hybrids between two cultivars and three wild species, the latter identified in earlier research as possessing resistance to late blight or PCN. Further work is needed, but the potential for genetic introgression via somatic fusion has clearly been demonstrated.

Molecular techniques, alluded to earlier as a means of increasing the efficiency of breeding using marker-aided selection, also provide extremely powerful tools for analysis and objective quantification of species relationships and biodiversity.

In a recent SCRI survey of 178 potato cultivars on the UK National List, using a combination of nuclear and chloroplast SSRs (microsatellites), a paucity of chloroplast genetic variation was highlighted, which was not seen at the nuclear level. Eighty-five percent (151) of these cultivars had exactly the same 'T type' cytoplasm. Our observations suggest that, unless diverse chloroplast types are actively chosen as parents in potato breeding schemes, the cultivated potato in the UK, perhaps Europe, even globally, may ultimately be represented by a single chloroplast haplotype. This could expose this extremely important food crop to the sort of devastating epidemic that occurred amongst the corn (maize) crop in the USA in 1970, when a strain of southern corn leaf blight attacked 70 percent of the maize in the USA, which had a single source of cytoplasm susceptible to the pathogen. An encouraging observation has been that the parents of the SCRI multitrait breeding scheme possess eight distinct cpSSR haplotypes compared to the two to three of modern and old cultivars respectively, a potentially serendipitous bonus that was not envisaged at the start of this scheme.

In addition to the research on potatoes, the Crop Genetics Department continues to maintain an interest in other crops. The commercially-funded swede breeding programme, based on single seed descent from F1 hybrids between selected SCRI parents, will reach the stage where the lines are sufficiently inbred for field trialling in 1999. Projects funded by the European Union and another by the Department for International Development are successfully achieving the efficient transformation of grain legumes and chickpea, and are described elsewhere in this year's report. The first summer's work on a MAFF-funded project, designed to quantify pollen and hence gene flow in oilseed rape, has produced some surprising – perhaps controversial – findings. Airborne pollen deposition was shown to decline steeply with distance to a low 'background' level, but a consistent fertilisation of about 5 percent of flowers on male sterile 'trap' plants was maintained up to 4 km from the nearest known fields of commercial oilseed rape crops. Though this may be an overestimate of what may occur with normal male fertile plants, DNA fingerprinting was able to confirm the source of pollen reaching the male sterile plants and that this was often a mixture of pollen from different source crops.

The commercially-funded potato breeding programmes continue to achieve, or exceed, their objectives. In addition to the two *phureja* clones mentioned above, three other *tuberosum* clones were also submitted to National List Trials, two being potential processing varieties from the Targeted Accelerated Breeding programme (Ann. Rep. 1996/97, 40-43). The new cultivars Amour (an export variety aimed at the Mediterranean region) and Blush (an attractive first early) were added to the National List and will be being commercialised by our private company partners.

Applied potato genetics and breeding: potato improvement by multitrait genotypic recurrent selection

J.E. Bradshaw, I.M. Chapman, M.F.B. Dale, G.R. Mackay, R.M. Solomon-Blackburn, M.S. Phillips, H.E. Stewart, G.E.L. Swan, D. Todd & R.N. Wilson

Introduction Today, in Britain, the most widely grown potato (*Solanum tuberosum* subsp. *tuberosum*) cultivars are susceptible to a range of pests and diseases, which have to be controlled by the widespread use of chemicals such as fungicides for late blight [*Phytophthora infestans* (Mont) de Bary], nematicides for potato cyst nematodes (PCN), and insecticides for aphid-transmitted virus diseases. However, chemical control is expensive, not always effective, and raises environmental and food safety concerns, particularly over large-scale insecticide use and pesticide residues in tubers for human consumption. Hence, new cultivars with higher levels of disease and pest resistance are highly desirable but, for commercial success, they must have acceptable marketable yields and meet the quality requirements of processors and supermarkets (Ann. Rep. 1997/98, 76-80).

Therefore, in 1991, we set up an experimental breeding programme designed to combine quantitative resistances to late blight and the white potato cyst nematode [*Globodera pallida* (Stone)] with commercially acceptable tuber yields and quality. Such resistances were available in *S. tuberosum* germplasm held at SCRI as a result of past breeding and introgression from wild and short day-adapted cultivated species, but had not been incorporated into widely grown cultivars. We decided to concentrate on quantitative field resistance to late blight because it has proved more durable than dominant R-gene resistance, which the fungus can readily overcome. We also decided to select for resistance in the tubers as well as in the foliage because the one does not guarantee the other and susceptible tubers can be infected by spores produced over a relatively long period of time from the slowly spreading sporulating lesions of a leaf-resistant cultivar. Whilst cultivars with major gene resistance (H1) to the common UK pathotype (Ro1) of the golden potato cyst nematode (*G. rostochiensis*) were being widely grown, only quantitative resistance was available to the common UK pathotype (Pa 2/3) of *G. pallida* (Ann. Rep. 1995, 30-34). It was also thought desirable to include in the breeding programme parents with virus resistance, particularly to Potato Leaf Roll Luteovirus (PLRV) and Potato Y Potyvirus (PVY). However, it was not possible to select for virus resistance during the programme, although the products of this research will be assessed for their resistance.

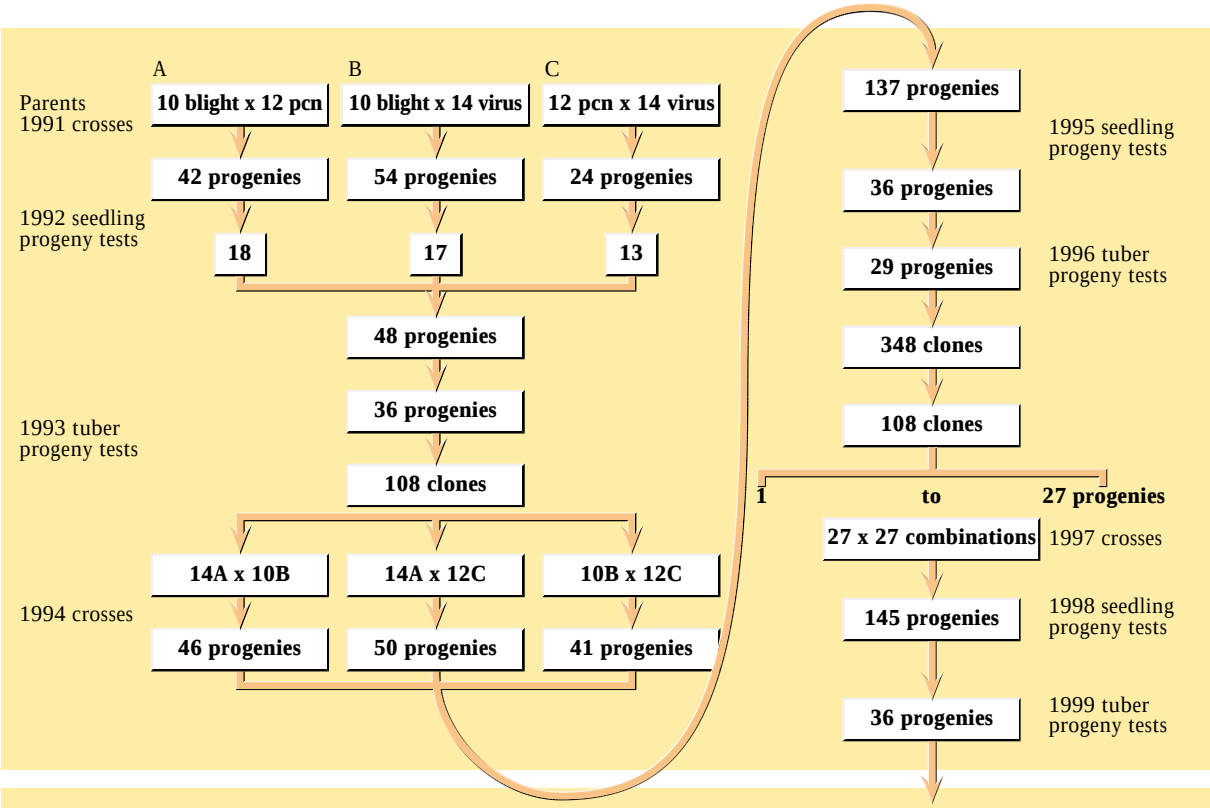

Figure 1 Multitrait genotypic recurrent selection programme.

The breeding programme had important research objectives because it was designed to use the seedling and tuber progeny tests developed at SCRI (see Bradshaw & Mackay[1] for review) for the first time in a multitrait genotypic recurrent selection programme. In genetical terms, the aim was to increase the frequency of desirable genes in the breeding material over a number of generations of recurrent selection, and in each generation to seek the best genotypes available for multiplication as new cultivars or parental material for future use. Clearly the shorter each cycle in years, the faster the overall rate of progress. In practice, we found that we could operate on a three-year cycle (Fig. 1) comprising crossing, seedling progeny tests, and tuber progeny tests each cycle.

Outline of the programme In order to combine desirable genes from three sets of parents, pair crosses were made in 1991 between the blight and PCN resisters (set A), the blight and virus resisters (set B), and the PCN and virus resisters (set C), followed in 1994 by crosses between progenies from different sets (AxB, AxC and BxC). Then, in 1997, crosses were attempted between the 27 progenies selected from the progeny tests. The overall success rate for crossing in 1991, 1994 and 1997 was 28%, 32% and 41% of desired combinations, figures that were typical for tetraploid potatoes.

In the 1992 seedling progeny tests, any progenies that were below average for breeders' visual assessment of tubers for commercial worth (breeders' preference) were eliminated, together with any that were below average for foliage blight and pcn resistance in set A, foliage blight resistance in set B, and PCN resistance in set C. In contrast, in 1995 and 1998, the best progenies were selected for further evaluation on the basis of a selection index, in which the breeders' preference, PCN and foliage blight scores in standard deviation units from their overall means were weighted by their heritabilities. Any progenies that subsequently proved too susceptible to tuber blight were also eliminated.

The best seedling progenies were grown in the field as tuber progenies at our high grade seed site (Blythbank Farm, Peeblesshire) in 1993 and 1996, and the same will happen in 1999. In 1993, two breeders visually selected the most attractive looking clone in each of three replicates for each of the 36 progenies to give 108 clones for the next round of crossing, as well as for evaluation as new cultivars. In 1996, a slightly different approach was taken. At harvest, the six most attractive looking clones from each of the two replicates of the 29 progenies were selected and tested for

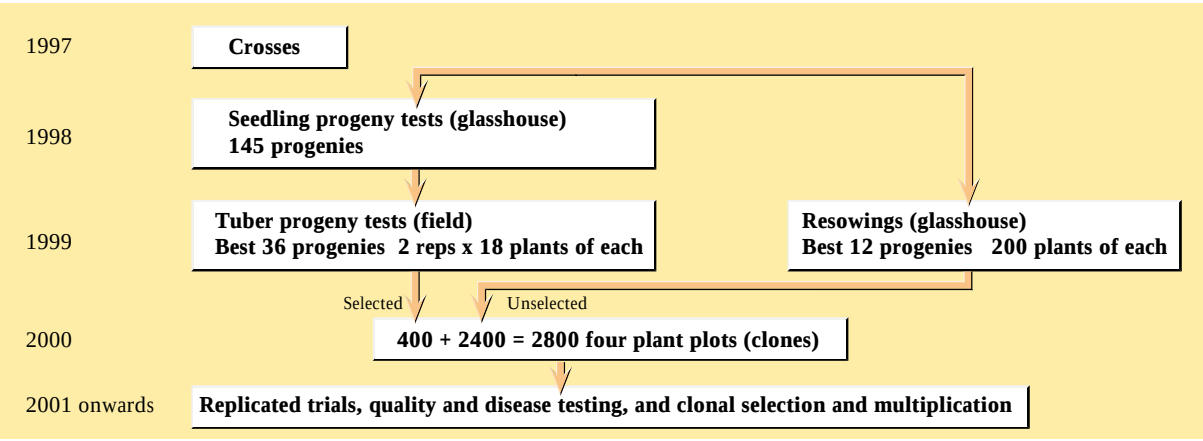

Figure 2 Cultivar production.

PCN resistance from 30 January to 16 April 1997. The 108 most resistant clones (cyst counts <40% of control cultivar Desiree) were then used as parents for the next round of crossing, as well as for evaluation as new cultivars. In 1999, after harvest, a tuber progeny test for fry colour is going to be introduced into the programme, as has already been successfully done in our targeted and accelerated breeding of potatoes for processing quality (Ann. Rep. 1996/97, 40-45). In other words, the multitrait breeding programme is an evolving research project.

More plants have been raised from the best crosses of each cycle to provide further clones for evaluation as potential cultivars. This year (1999), for example, 200 true seeds from each of 12 of the best progenies from the 1998 seedling progeny tests have been sown in a glasshouse (resowings) to provide up to 2,400 clones for visual assessment in four-plant plots at our seed farm in the year 2000 (Fig. 2). The selected clones will then enter a multistage multiplication and selection programme involving replicated yield trials and quality and disease testing, as described by Bradshaw and Mackay[1], in order to provide potential cultivars to private companies for multisite trials and submission to National List Trials.

Progress to date An indication of our progress to date is given by the results of the 1998 seedling progeny tests (Fig. 3). There was enough seed for two replicates of 25 seeds of 145 progenies for each of the breeders' preference, PCN and foliage blight progeny tests, and of 122 progenies for the tuber blight progeny test. Breeders' preference is the only progeny test where individual clones (2 x 18) within progenies are assessed and a mean taken. This is done by at least two breeders on a 1 to 9 scale of increasing preference, where 3 or less is unacceptable and 5 or more is acceptable, with 4 borderline.

The broad sense heritabilities (h^2_B, proportion of variation which was genetical) ranged from 0.39 for foliage blight to 0.82 for tuber blight, and were similar to those found in 1992 and 1995, except for foliage blight, which was slightly lower (0.55 in 1992 and 0.63 in 1995). The highest correlation between traits was the one between resistance to foliage blight and resistance to tuber blight, which was statistically significant (P<0.1%) but small in magnitude (r=0.35), and underlined the need to select for resistance to both in a breeding programme.

The 12 progenies selected for resowings in 1999 were chosen using a selection index based on Smith's[2] discriminant function for plant selection with relative

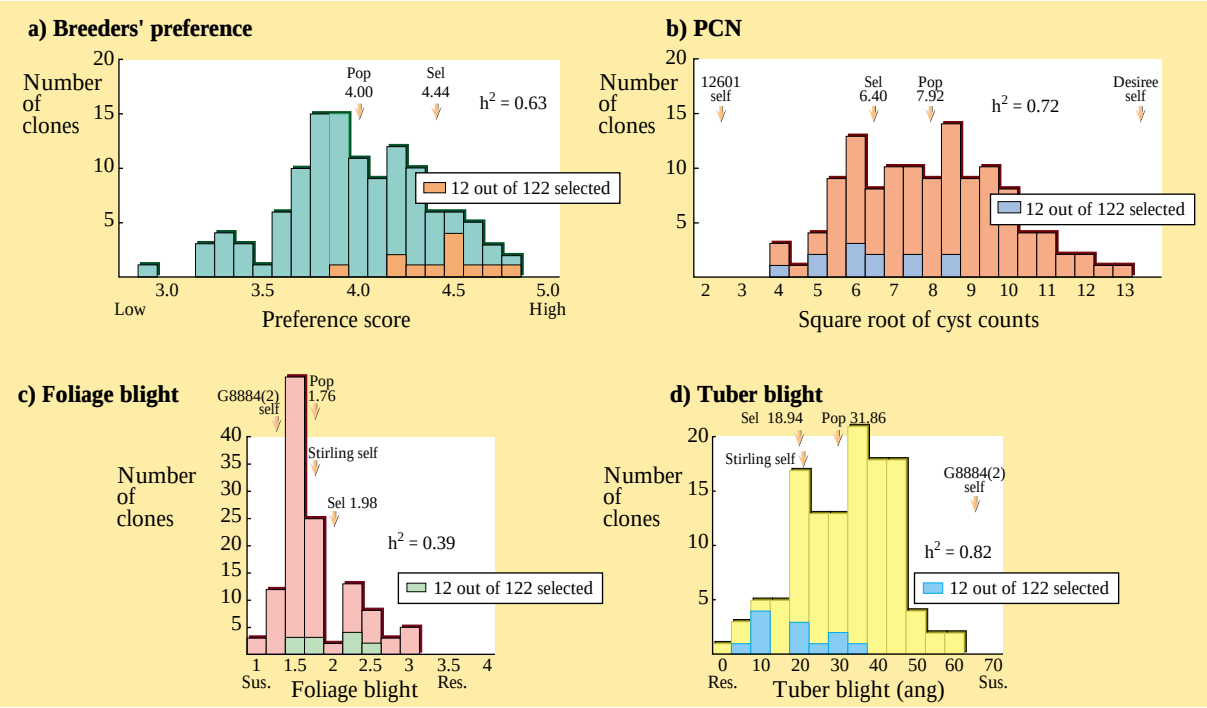

Figure 3 1998 progeny tests for a) breeders' preference; b) PCN; c) foliage blight; d) tuber blight.

economic weights in phenotypic standard deviation units of 1:1:½:½ for the four traits, so that foliage and tuber blight together were given a similar weight to breeders' preference and pcn. The mean of the 12 progenies was 4.44 for breeders' preference (individual clones:<4 reject, ≥5 definitely select); 6.40 for PCN, which was closer to the resistant than the susceptible control; 1.98 for foliage blight, which was better than the resistant control; and 18.94 for tuber blight, which was as good as the resistant control. Hence, in seeking new cultivars and parents from these progenies, there is a good chance of finding clones that are better than these means for all four traits, but it remains to be seen if they are as good as the best of the original parents for individual traits. This critical assessment will come in the year 2001, when the best clones from the 12 progenies are compared with the parents used in the original crosses.

Diallel analysis of variation in population The genetical variation in the multitrait breeding population has been evaluated through a 15 x 15 diallel set of crosses, including selfs and some reciprocal crosses, made in 1992[3] and subsequently assessed in seedling and tuber progeny tests. Five male fertile parents were chosen from each of the three sets of parents used in 1991. The large amount of GCA (General Combining Ability) variation found for breeders'preference, PCN, foliage blight and tuber blight was indicative of much additive genetic variance in the population. Furthermore, the only statistically significant ($P<0.05$) correlation between GCAs for different traits was a favourable one ($r=0.56$) between foliage and tuber resistance to late blight. It was concluded that prospects were good for simultaneously improving all four traits over a number of generations of multitrait genotypic recurrent selection. It was also concluded that the variation due to SCA (Specific Combining Ability), which was found for breeders' preference and tuber blight, could be exploited in selecting the best crosses for cultivar production. The assessment of other economically important traits, including yield and fry colour, was done on tuber progenies at our high grade seed site from 1994 to 1996, and in replicated ware trials in 1995 and 1996. Fry colour had the highest narrow sense heritability ($h_n^2 = 0.90$), measured as twice the GCA variance divided by the phenotypic variance (i.e. observed variation) of progeny means. Therefore, the 36 tuber progenies being evaluated in the breeding programme in 1999 will be selected for this important processing characteristic, as already mentioned. Emergence, maturity, yield, dry matter content and dormancy were identified as other traits that would respond to progeny (i.e. full-sib family) selection.

Future possibilities Although the diallel analyses provided valuable information about the genetic varia-

tion present in the multitrait programme for traits displaying continuous variation, they did not provide any clues as to the number of genes segregating, their chromosomal locations, or what they do. Therefore, two of the crosses from the diallel were chosen for a more detailed genetical analysis using molecular markers in collaboration with Robbie Waugh and his colleagues in the Potato Genomics Unit and with Christine Hackett in BioSS. AFLP markers have already been identified which are associated with resistance to foliage blight in cv. Stirling, resistance to *G. pallida* in clone 12601ab1 (ex. *S. tuberosum* subsp. *andigena*) and resistance to *G. pallida* in clone 12288af23 (ex. *S. vernei*), and which account for between 20% and 34% of the phenotypic variation. Hence, we can start to explore whether or not molecular marker assisted selection can improve the efficiency of the multitrait breeding programme, both for selecting parents for the next round of crossing, and for identifying superior genotypes within the best progenies at an earlier stage than could otherwise be done.

As the European potato is a tetraploid, which displays tetrasomic inheritance, it is also worth exploring whether or not faster progress is possible by haploidisation to the diploid level, followed by recurrent selection at the diploid level, before polyploidisation back to the tetraploid level for cultivar production. However, this will have to await the outcome of current research on microspore embryogenesis, because a much larger number of both male and female fertile dihaploids are required than has proved possible by the standard *S. phureja* inducer method.

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Barley domestication – *Hordeum spontaneum*, a source of new genes for crop improvement

R.P. Ellis, J. Russell, L. Ramsay, R. Waugh & W. Powell

The challenge that faces farmers in the future is the sustainable production of crops for an expanding human population. It is possible to envisage a number of pressure points such as the need for improved resistance to pests and diseases, greater fertilizer efficiency and economic water usage. In particular circumstances, it may be necessary to develop farming systems that use brackish water. The breeding of crops for such complex objectives requires the development and use of marker-assisted breeding schemes. But before marker-assisted breeding schemes can be implemented, gene maps are required to locate suitable genes.

To date, our use of molecular markers has focused on their development for use in breeding and for gene isolation for transformation (SCRI Ann. Rep. 1997/98, 64-66). One other important area of research in plant breeding is the discovery of new genes or alleles, particularly in the wild relatives of crop species. Our work with wild barley has appeared in a number of reports (SCRI Ann. Rep. 1992, 20-23; 1993, 39-40) and a cross between a cultivar and wild barley was particularly useful for gene mapping, as it provided wider contrasts (SCRI Ann. Rep. 1996/7, 82-84) than crosses between cultivars (SCRI Ann. Rep. 1995, 59-62).

Wild barley exists as isolated populations throughout the Eastern Mediterranean, Middle East, and Northern Asia (Fig. 1). Our concept of wild barley and its relationship to cultivars depends on the systems used for collection and assessment. It has not been possible to use markers to genotype material as it is collected, so the main strategy has been to sample populations systemically. This implies that collections that exist in many centres, e.g. John Innes Centre, Norwich and Institut für Pflanzengenetik und Kulturpflanzenforschung, Gatersleben, Germany, will contain a wide range of genotypes but are completely uncharacterised. This has hampered the utilisation of the material for breeding, and in the past its main use has

Figure 1 The Middle East, particularly the Fertile Crescent (modern day Israel, Lebanon, Syria and Iraq with Turkey and Iran) has been defined as the Centre of Origin of many cereal crops and is the region in which wild barley (*Hordeum spontaneum*) shows greatest variability.

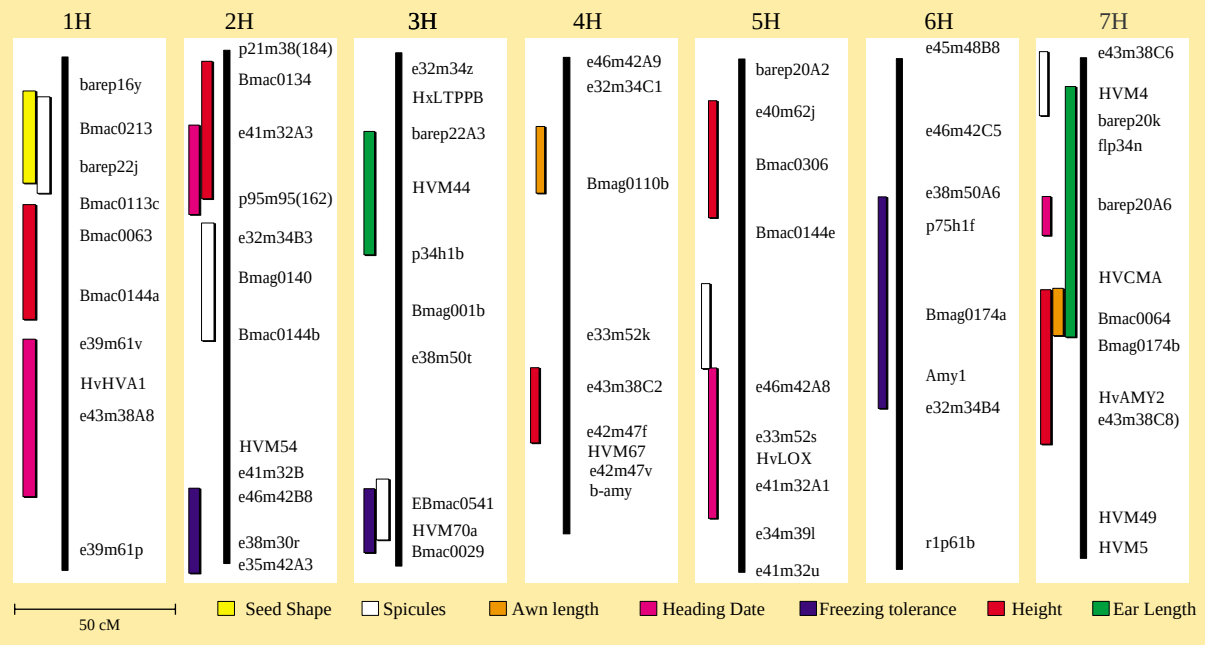
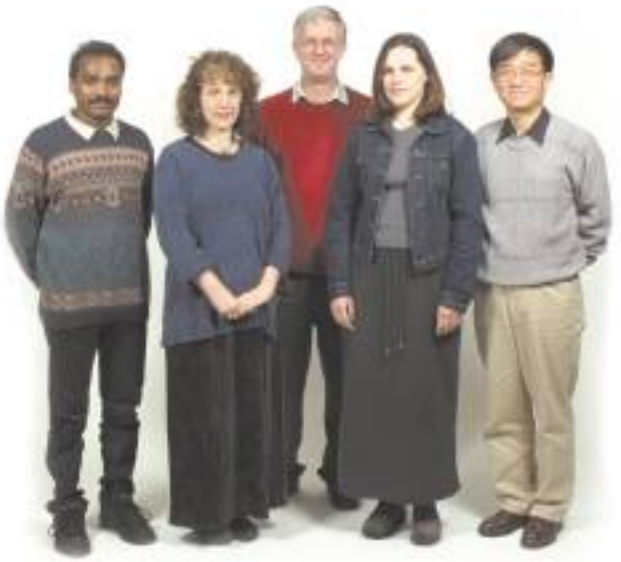


Figure 2 Genome map locating quantitative traits related to domestication in the cross Lina x HS92.

been for the improvement of easily selected characters such as disease resistance.

Wild barley strongly contrasts with cultivars for a number of characteristics. Typically, in wild barley the lemma awns are much more barbed, are tougher and the ear shatters when ripe. These are adaptations to animal dispersal and not suited to cultivation. Relatively high levels of dormancy in wild barley often prevent immediate germination, and time of flowering depends on the satisfaction of a high vernalization requirement and short-day photo-period response. Again, these are not desirable characteristics in cultivars. However, when a simple selection scheme exists, as in the case of mildew resistance, the use of back-crossing often fails to transfer single genes and a whole chromosome segment ends up in the progeny. One common result is the association of disease resistances with characters that impede the fullest development of malting quality (SCRI Ann. Rep. 1993, 24).



The processes of domestication have resulted in similar changes in all cereal species, so that wheat, rye and barley cultivars all have non-shattering ears and flower at a time that is suitable for crop production. Our research has provided an understanding of the contrasts between wild barley and cultivars for characters such as height, time of flowering, ear size and the spiculation or barbing of the grain (Fig. 2). Time of flowering and height are related to genetic mechanisms that control plant development and growth. In turn, these result from genes that control physiological characters, essentially the rate of cell division and cell size. This is clearly seen in analyses of the effects of genes that give semi-dwarf plant stature. There is a contrast between cultivars that possess the *ari-e*.GP gene on chromosome 5H (SCRI Ann. Rep. 1993, 30) and those with the *sdw1* gene on chromosome 3. The former are tolerant to salt while the latter were less tolerant in these tests. The reasons for this contrast in tolerance are not know. The *ari-e*.GP gene was found in the successful Scottish cultivar Golden Promise and was associated with early flower-

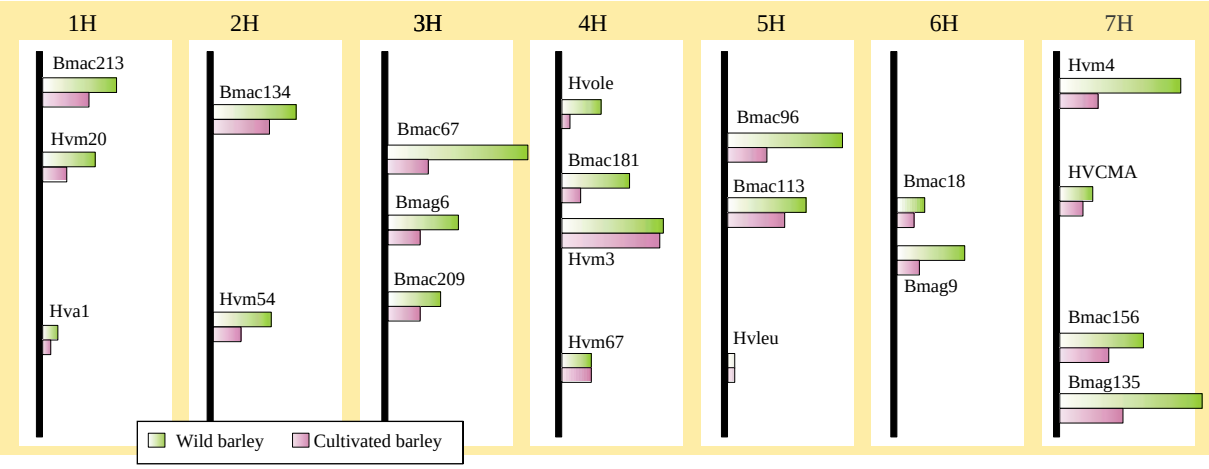


Figure 3 Allelic variability present in wild barley compared to cultivated barley during domestication and selection.

ing and low yield potential. More modern cultivars, such as Cooper, Landlord and Optic, are later and have much higher yield potential, essentially due to larger grain size than the cultivars with *ari-e*.GP. The contrast between cultivars and wild barley is even more extreme with a wide range of flowering dates and very small grain size, i.e. a relatively smaller investment of limited resources into reproduction as numbers of seed matters more than size resulting in greater tolerance of abiotic stresses.

In addition to revealing the current genetic composition of cultivars, our studies have revealed the effects of selection during the process of domestication. Some very obvious differences between wild and cultivated barley, for example the appearance of a tough ear, reduction in awn stiffness and an increase in grain size, must have occurred very rapidly. Other differences, such as increases in grain yield and malting quality, have been accumulated over successive cycles of selection in breeding programmes. While it was expected that height and heading date would show more than one quantitative trait locus, it was more of a surprise to find that lemma spicules were determined by five QTLs. Spicules on the lemma have been used as a diagnostic trait for the identification of cultivars but modern spring barleys have relatively few. In contrast some winter barleys, e.g. Malta and all wild barleys, have highly spiculate lemmas. The origin of smooth skinned barleys is in the hand-evaluation methods used to select malting barley cultivars, prior to the development of micromalting at the Plant Breeding Institute, Cambridge, in the 1960s. The result of hand-evaluation was that malting cultivars of the 1950-1980 period were lower yielding than feed types. At the time, this was attributed to the greater complexity of selection for malting quality than feeding types. Our new evidence suggests that the root cause of the problem is the simultaneous selection for alleles at a number of loci. This selection may have acted directly or indirectly to reduce plant vigour. In this context, it is interesting to note that silicon, a respiratory irritant in grain dust, is an important plant nutrient that is essential for plant structure and disease resistance. This is very apparent in hydroponic experiments, where the omission of silicon results in an increase in disease susceptibility.

Our experience in mapping genes of economic importance (SCRI Ann. Rep. 1994, 60) shows that very dense genetic marker maps are needed to analyse the complex effects of important genes such as *ari-e*.GP or *sdw*1. It is still difficult to separate the direct effects of these genes from indirect effects, or the effects of distinct but closely linked genes, without obtaining a complete gene sequence and determining gene function. For the present, our work has concentrated on setting the scene by a retrospective analysis of the genetic composition of cultivars since 1880.

The cultivars sampled in this study include genotypes from the 1880s through to modern-day cultivars. This affords an opportunity to examine changes in levels and patterns of variability over time and establish, in a quantitative manner, whether the genetic base of barley is being eroded. By representing the distribution of alleles in the form of a two-dimensional graph (Fig. 3), we can begin to highlight specific allele substitution events and examine the effects of selection over time. Each allele is coded with a different colour and loci are ordered linearly into linkage groups. The overall effect is a reduction in colours as we move from the top to the bottom of the graph, suggesting a loss of alleles

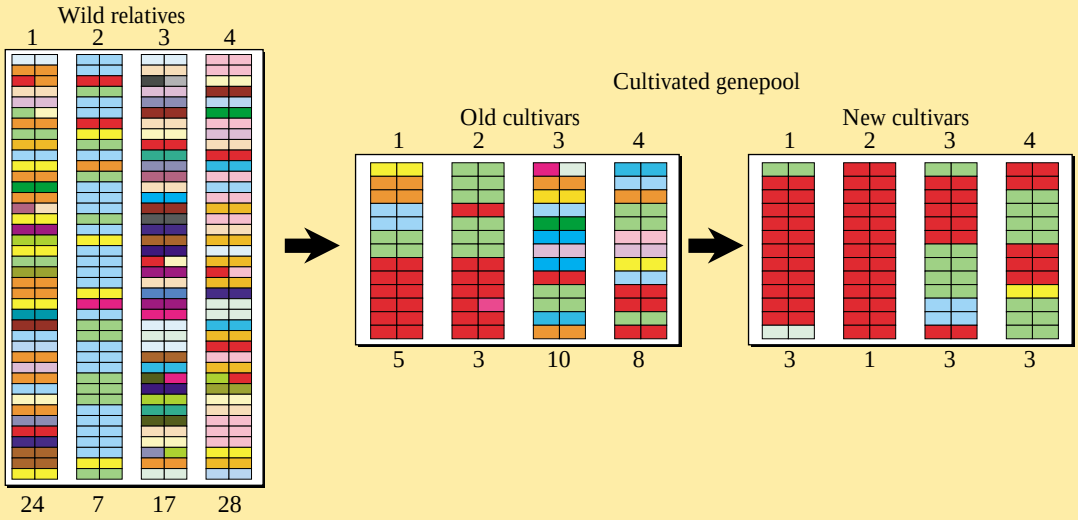


Figure 4 Allelic composition of wild barley compared to cultivated barley on chromosome 7H. The SSRs are coded (1) Hvm4, (2) HvCMA, (3) Bmag135, (4) Bmac156. The numbers of distinct alleles found in wild barley, old cultivars and new cultivars are given at the foot of each column.

over time. These results indicate clearly that the effect of commercial plant breeding, with the need to demonstrate distinctness, uniformity and stability before cultivar registration, was to produce crops that were more genetically uniform.

Partitioning of variation between wild and cultivated barley has re-emphasised the comparatively narrow genetic base of barley, with 54% of the alleles being unique to wild barley and absent in the sample of cultivated genotypes assayed. Only 39% of the alleles in the wild gene pool have been transmitted into the cultivated gene pool. An examination of the distribution of allele frequencies in *H. spontaneum* and *H. vulgare* confirms the loss of rare alleles and decrease in genetic diversity during domestication, with a further narrowing of the genetic base by subsequent breeding (Fig. 4).

Comparisons between the gene pools provides evidence of successive genetic bottlenecks in the domestication of barley as well as in the development of the most recent cultivars. The constraints imposed by lack of genetic variation in cultivated barley can be overcome by exploiting molecular markers to access a broader spectrum of genetic variation from wild barley. In this way, molecular markers, particularly microsatellites, have a significant role in promoting the sustainable use of genetic resources.

Potato genomics: a general strategy for the molecular genetic characterisation of *Solanum* germplasm

G. Bryan, W. De Jong, J. Provan, D. Milbourne, J. McNicoll, J. Davidson, G. Ramsay, R. Waugh

In recent years, a major goal of potato genetic research at SCRI has been the development of PCR-based molecular genetic markers. The primary use of these markers has been for the analysis of segregating populations of potato, which has allowed the genetic locations of markers to be determined. In combination with phenotypic data obtained from such populations, it is also possible to identify molecular markers linked to traits of biological and agronomic interest. A further objective is to use these powerful genetic markers for further analysis of populations and the genetic characterisation of potato germplasm. This germplasm may take the form of tetraploid potato cultivars and breeding lines, as well as accessions of wild and primitive cultivated potato species of various ploidy levels, such as material from the Commonwealth Potato Collection (CPC).

Simple Sequence Repeats (SSRs)

At the present time, approximately 150 microsatellite or simple sequence repeat (SSR) loci have been identified and characterised in potato at SCRI. Progress in mapping and characterising these SSRs has been reported in previous Annual Reports (e.g. Ann. Rep. 1997/8, 86-88). Microsatellites offer the advantage of showing a codominant mode of inheritance, meaning that several alleles can be identified for each locus, subject to the degree of polymorphism shown by that locus. This property is particularly useful in tetraploids, and allows the possibility of assigning microsatellite genotypes (the combination of alleles present at a set of SSR loci) for any given accession. SSR markers have been used very effectively to anchor genetic maps containing large numbers of high-volume, 'single-dose', markers such as Amplified Fragment Length Polymorphisms (AFLP). AFLP technology has become the molecular workhorse for the generation of high volume markers in potato genetic mapping. AFLP markers typically exhibit a 'dominant' (i.e. presence/absence) pattern of inheritance that makes the establishment of genetic maps based on AFLPs alone somewhat problematical, particularly in tetraploids. Incorporation of co-dominant SSR marker locus data greatly reduces the problem of map construction by, for example, allowing the rapid assignation of linkage groups to chromosomes. Microsatellite markers are eminently suitable for the analysis of potato germplasm, from cultivars and breeding material to wild species.

Chloroplast SSRs

In recent years, SCRI has led the way in the development of microsatellite markers from chloroplast genomes of several plant species, including maize and pine (Ann. Rep. 96/97, 93-4). Chloroplast genome-based microsatellite markers are attractive in that they are easy to deploy (owing to the high copy number of the cpDNA molecule per cell) and, due to the lack of recombination in the chloroplast genome, it is very straightforward to assign a cpSSR 'haplotype' to any given accession. We have identified a set of 36 primer pairs, which amplify polymorphic PCR products from the chloroplast genomes of most Solanaceous plants, including potato and tobacco. The origin of these primers is the completely sequenced tobacco chloroplast genome, but the chloroplast genome sequence is sufficiently conserved among Solanaceous plants to permit cross-species PCR amplification. Figure 1 shows an example of a cpSSR locus PCR assay on a set of potato germplasm from the

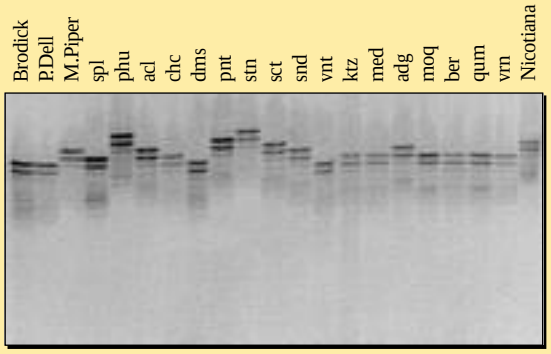


Figure 1 Chloroplast microsatellite NTCP12 applied to a diverse set of potato/tobacco germplasm.

CPC. Figure 2 provides information on levels of polymorphism obtained with some of the more polymorphic cpSSR primer pairs. These primers almost certainly will prove useful in all Solanaceous plant species. Owing to the ease of deployment of these markers, it is possible to 'multiplex' several cpSSR primer pairs in a single PCR reaction, meaning that a cpSSR haplotype can be obtained using just a few PCR reactions. These markers will have several applications, such as examining the levels of chloroplast genome variation in the cultivated potato gene pool (see below), as well as for phylogenetic and diversity analysis of potato germplasm.

Deployment of these markers in the analysis of potato germplasm:

1. SURVEY OF POTATO CULTIVARS AND COMMONWEALTH POTATO COLLECTION In order to assess their utility for germplasm analysis, the 36 cpSSR primer pairs described above were applied to the analysis of a representative set of 25 wild species from the CPC and a set of 30 potato cultivars. The cultivars were selected to represent a wide range of chloroplast diversity; these had been identified in a previous study, based on chloroplast derived RFLPs. Our preliminary survey, using these 36 primer pairs, showed that 26 (72%) detected some level of polymorphism among the 55 accessions. This study showed that the cpSSRs are very suitable for the analysis of potato and tobacco germplasm at several taxonomic levels[1]. A cluster analysis of material from the CPC shows good agreement with previously published phylogenies of potato. Secondly, it appears that amongst the diverse set of potato cultivars, there is a predominant 'cytotype', which is thought to result from the widespread use of the cultivar Rough Purple Chili as a female parent in the latter half of the 19th century (Fig. 3). A third observation is that some cultivars and breeding lines

Locus	Av. Heterozygosity	No. of alleles
NTCP3	0.59	4
NTCP23	0.66	5
NTCP4	0.62	4
NTCP6	0.59	7
NTCP7	0.58	3
NTCP8	0.66	5
NTCP9	0.70	8
NTCP30	0.54	4
NTCP12	0.58	5
NTCP14	0.69	5
NTCP18	0.66	3
NTCP39	0.62	4

Figure 2 Levels of polymorphism among 12 of the most polymorphic cpSSR loci. Average heterozygosity ($\hat{H}$) has been calculated as follows : $\hat{H} = n/n\text{-}1(1\text{-}\Sigma p_i^2)$
where p_i = frequency of i^{th} allele
n = number of samples

have a cpSSR haplotype identical to that of particular wild species of potato, which were used in the past to introgress genes of agronomic importance into cultivated *S. tuberosum*. This latter observation suggests that a large part of the chloroplast diversity still present in cultivated potato is derived from wild or primitive cultivated species.

2. UK POTATO NATIONAL LIST SURVEY A combination of nuclear SSRs and cpSSRs has been applied to the analysis of 178 potato cultivars on the UK

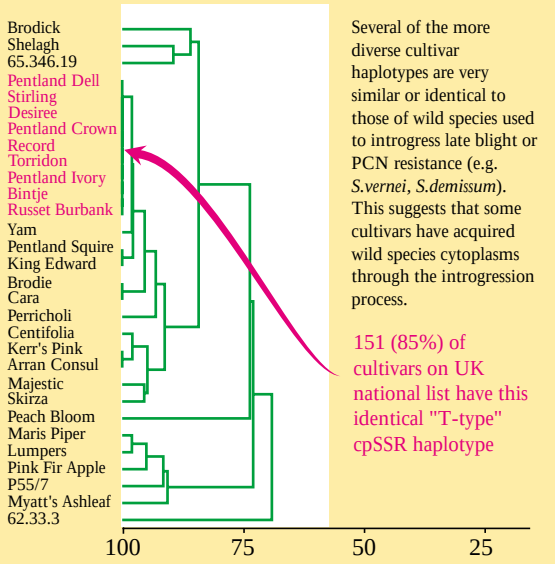


Figure 3 Cluster analysis of chloroplast haplotypes of 30 potato cultivars. Haplotypes have been clustered using a UPGMA method based on similarity estimates obtained using a 'city-block' method.

National List. This study has highlighted a paucity of chloroplast genetic variability in the cultivated potato in Europe, which is not seen at the nuclear level[2]. A very large proportion of these cultivars (151 i.e. 85%) has exactly the same type of 'T-type' cytoplasm described above. The high levels of nuclear SSR variability do not differ markedly among different cpSSR haplotypes and, moreover, the large group with identical cytoplasms shows no less nuclear polymorphism than the remaining cultivars. These observations suggest that, unless diverse chloroplast types are actively chosen as parents in potato breeding schemes, there is a strong possibility that the cultivated potato in the UK, and possibly Europe, will ultimately be represented by a single chloroplast haplotype.

3. MULTITRAIT RECURRENT SELECTION SCHEME An analysis of genetic variation among 23 of the parental lines used in a multitrait recurrent breeding scheme has been initiated. To provide a comparison, representative samples of 24 old cultivars (pre-1900) and 20 popular cultivars currently grown in the UK have been included in this study. These accessions have been analysed with nuclear and chloroplast SSRs as well as AFLP markers. AFLP and nuclear SSR analysis show that the parents of the multitrait scheme and, to a lesser extent, modern cultivars, show significantly greater numbers of 'rare alleles' than the old cultivars (Fig. 4). It is also clear that only a small number of rare alleles, present in the older material, have been lost from the cultivated potato genepool. A very encouraging observation from this study, provided by cpSSR analysis, is that the parents of the multitrait breeding scheme are represented by the presence of 8 distinct cpSSR haplotypes. This compares with two and three haplotypes in modern and old sets of cultivars respectively. The potato breeding industry as a whole should be encouraged to adopt a similar strategy in the choice of parents for potato breeding schemes.

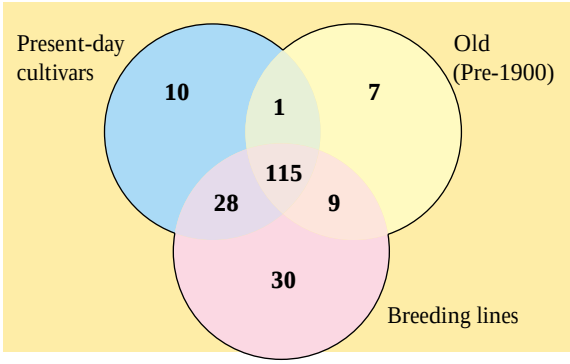


Figure 4 Distribution of polymorphic AFLP markers among three sets of adapted potato germplasm.

4. CPC SURVEY In the last year, we have started to use microsatellite markers to examine additional material from the CPC. These studies were designed to test further the utility of SSRs for the analysis of potato germplasm across relatively wide genetic distances. For example, a set of nuclear and chloroplast SSRs was used to study 33 accessions from series Longipedicellata, which contains several species. This study has indicated that there appear to be no distinct boundaries between groups of accessions representing different species from this complex, suggesting that the classification into separate species should be re-examined. Recently, we have initiated a collaboration with Dr David Spooner (University of Wisconsin, USA), who is currently examining accessions from series Longipedicellata, to carry out an extended molecular and morphological analysis of material from this complex. This work has implications for potato taxonomists and for the maintenance of larger germplasm collections, such as the CPC, where the accurate identification of accessions is highly important, particularly when the costs of preserving such collections are considered. A second study, recently initiated, is to examine potential progenitor species of cultivated potato, in an

attempt to elucidate further the likely origins of the cultivated potato.

Potato germplasm evaluation - a general strategy

Microsatellites remain the recommended type of marker for the examination of potato germplasm, owing to their ability to detect allelic variants and high levels of genetic polymorphism. The need for examining chloroplast DNA variation independently of the nucleus is highlighted by the results outlined above. Differences in nuclear genotypes may not preclude identical cytoplasms, although accessions with different cytoplasms are very unlikely to have the same nuclear constitution. This suggests that a sensible strategy may be to deploy cpSSRs as a primary screen for polymorphism. If higher numbers of markers are required, say for fingerprinting more closely related accessions, the use of AFLPs may also be an option. For most purposes, however, the combination of the 10 most polymorphic cpSSRs in combination with 10-20 nuclear SSRs will be sufficient.

The future

We will continue to use molecular markers for the analysis of potato germplasm at SCRI. Further studies will involve adapted material, such as potato cultivars and breeding lines, segregating populations, and material from the CPC. Ultimately, we would like to genetically 'fingerprint' the entire CPC. This would provide a much stronger basis for the use of the CPC as a source of novel genes (e.g. disease resistance). It would also provide a stronger means for a rational framework on which to maintain the collection. This would be of even greater benefit if other large potato collections (e.g. CIP, Peru) could be examined with the same genetic markers. This would permit quantifiable assessments of the relative levels of genetic diversity within and between different germplasm collections.

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Cultivar responses to long-cane fruit production in raspberry

T. Gillespie, R. Brennan & R.J. McNicol

Introduction Currently the United Kingdom is a net importer of raspberries. Traditionally the main outdoor raspberry cropping season in the UK begins in June with summer fruiting cultivars and is extended through to late August early September by the use of primocane/autumn fruiting cultivars.

In recent years, consumer demand for fresh raspberries outwith the main production season has increased, with high premiums being paid for fresh market raspberries. This demand is being met by imports from countries such as Spain, Portugal and Chile, which have the advantage of more favourable weather conditions and a longer growing season. Production of raspberries for fresh market either side of the main season is achieved through protected cropping under polytunnels, or in some cases glass, using novel systems of production to manipulate flowering and fruiting. Protected cropping and out-of season production in European countries is expanding, so that in areas of southern Spain nearly 100% of fresh market dessert raspberries for early, main and late season are being grown under tunnels.

The economic advantage and current consumer demand for out-of-season raspberry production has led growers in the UK to try new production techniques for extending the cropping period. One production technique, termed 'long-cane production', is increasingly being adopted, especially in protected cropping situations.

Long-cane production This system involves the manipulation of summer fruiting cultivars by cold storage of the canes. Long-canes are produced in the field in spawn beds. In autumn, when the canes have matured and are dormant, they are lifted from the beds with roots and placed into cold store. When adequate chilling has been accumulated, the long-canes are brought out of the cold store and planted for cropping under the protection of either polytunnels or glass. Production can either precede the main season, by removing the canes from storage as soon as dormancy has been satisfied, or be delayed by leaving the canes in the cold store until later in the season. After fruiting the mature canes are cut back and new spawn is allowed to grow for fruiting in the following year. The new spawn can either be incorporated into an annual or biennial plantation system, or lifted from the fruiting bed and cold stored again.

In truth, the success of long-cane techniques under protected cropping in the UK has been mixed, particularly regarding bud break and cropping consistency. Greater understanding of the plant has been required to improve the reliability of these production systems.

Research at SCRI This article describes research being carried out at SCRI on the long-cane production system. The ultimate goal of the 'Year-round soft fruit production' project, funded by MAFF (Project Number HH1519TSF), is to develop a commercial blueprint for the production of out-of-season *Rubus* crops in the United Kingdom. Commercial trialling in the project is carried out in conjunction with ADAS and in close collaboration with local propagators.

To improve the competitiveness of the long-cane system, research has been aimed at examining various means of optimising every stage of production, through a better understanding of the physiological processes involved with the aim of maximising fruiting potential. The main areas of long-cane production being studied are:

- Cane production
- Cold storage
- Dormancy and flower initiation
- Crop production

Figure 1 Traditional spawn bed production of primocanes.

Cane production The growing of canes for protected cropping in the following year has several specialised aspects. Currently, in the commercial sector, long-cane primocanes are being produced in traditional spawn beds (Fig. 1). The density of the growth within traditional spawn beds has clear effects on the quality of the cane produced, in terms of morphology and development. For example, canes within the dense canopy tend to initiate flower primordia *ca*. 2 weeks later than canes at the outside of the bed[1]. The quality, quantity and temporal distribution of light are known to influence correlative inhibition (apical dominance) and therefore plant growth and morphology[2]. A trial to examine the effect of initial spawn bed planting density and subsequent light penetration on root size, bud break and flower production is in progress.

Figure 2 The effect of infection by *Botrytis cineria* in cold stored canes.

In addition, the time of lifting canes from the spawn bed is critical: canes lifted too early show poor survival in cold stores even if the storage temperatures are reduced slowly. However, delay in lifting can leave canes vulnerable to frost injury with impaired cropping in the following year[3].

Lifting date has been found to have a significant effect on cane survival post-storage. Survival of cultivars Glen Moy and Glen Clova lifted before October was found to range from 0-16 %, from October survival post-storage increased to 80%.

Cold storage The ability of raspberry canes to store in a viable condition is crucial to the success of the long-cane system, with regard to meeting the chilling requirement of the plant, preventing growth, desiccation and disease while in the store, and avoiding low-temperature injury.

Cold storage of raspberry canes is necessary to fulfil the chilling requirement of the canes after lifting from spawn beds in the autumn. Storage of canes at 4ºC for 6 weeks is sufficient to meet the chilling requirement of upper buds in Glen Moy. The intensity dormancy attains can be influenced by environmental conditions, the age of the plant and cultivar differences[4]. Other factors, including the woodiness of the stem and the water content of both the bud and stem may also affect dormancy and chilling requirement. The deeper the dormancy attained, the more chilling is required for bud break, and therefore the minimum amount of chilling required to break dormancy cannot be regarded as constant for any one cultivar.

Fruiting potential and cane quality must be maintained through cold storage. Storage conditions must prevent bud desiccation and reduction in cane quality in storage. The major problems in storage to date have been desiccation of the cane and disease problems, predominately cane *Botrytis* infection. Comparison of bare root and covered root has established that bare root cane will desiccate in cold storage. Desiccation of buds is also an important consideration. Breathable fleece has been used to wrap around bundles of canes to maintain humidity and defoliation

treatments have also been included in these storage trials. A higher incidence of *Botrytis* infection and cane death (Fig. 2) was observed when fleece was used and leaves were left intact on the canes.

For long-term storage (greater than 4 months) it is apparent from our present studies that temperatures lower than 4ºC are necessary to prevent bud break and growth in storage. Canes of Glen Lyon that had been stored at 4ºC with 24 hour dark began to break bud and etiolated laterals developed after 4 months in storage[5].

Canes of cv. Glen Ample have been successfully stored for up to 10 months post-lifting. For long-term storage, 1ºC was found to be necessary to check bud break and growth. When grown on under protection, these canes produced a crop from late October through to mid November. Long-term storage was found to significantly decrease the time from bud break to flowering, from approximately 11 weeks (under normal field conditions) to just under 4 weeks. This response has also been observed in peach[6], where it appeared that further chilling caused compositional changes, particularly in the cell membranes, and produced a more concerted bud break.

Cane physiology In order to manipulate fruiting of raspberry outwith the main summer season, dormancy status within the over-wintering (resting) bud and the developmental stage of the apical meristem (i.e. vegetative or reproductive) must be accurately assessed. To obtain the maximum fruiting potential from cold stored canes, buds should have passed through endo-dormancy (true winter dormancy) and be in an eco-dormant (environmentally imposed) state, and flower primordia should be fully differentiated.

Flower initiation (Fig. 3 and Fig. 4) and development of dormancy in raspberry occur concurrently but independently in response to the lower temperatures and shortening daylengths of autumn. When buds become *endo*-dormant, their water content is reduced as free water becomes bound to hydrophilic proteins (macro molecules) in cell membranes[7]. A period of chilling is then required to break dormancy, leading to an increase in free water.

Work with S. Glidewell (CEP Dept.), using Nuclear Magnetic Resonance Imaging (NMR), and I. Roberts (Virology Dept.), using Confocal Laser Scanning Microscopy (CLSM) is being carried out to develop novel techniques for monitoring the water status and flower primordia development of raspberry buds.

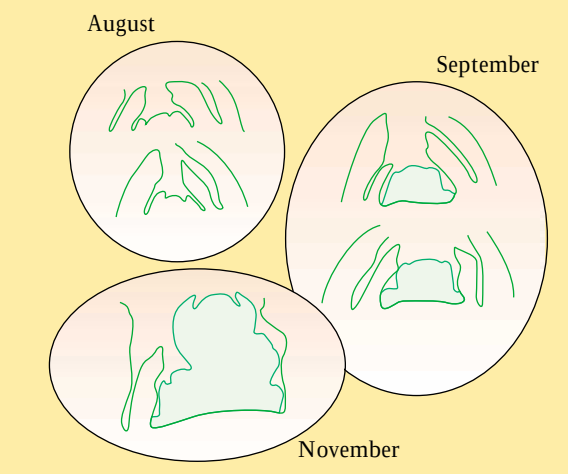

Figure 3 Schematic representation of floral development in raspberry (after Williams, 1950). The apical meristem changes from vegetative to reproductive in response to shortening days. The shape of the apex is modified and becomes domed and conical as the flower primordia develop.

NMR micro-imaging represents a non-invasive approach for evaluating changes in the behaviour of water in biological systems[8]. The basic principles of NMR spectroscopy and 3-dimensional imaging have been previously described[9,10].

Using NMR micro-imaging, we have been able to monitor changes in the water status of the same raspberry bud from the point at which it entered dormancy in the autumn through to bud break in the spring. NMR imaging is performed with a Brucker AM300/WBFT spectrometer (7.1 Tesla). A 20 mm coil has been modified to accept a woody plant speci-

Figure 4 Median longitudinal section of a bud taken with a Zeiss Tessovar macro camera. Terminal and axillary flowers can be seen developing at the centre of the bud.

Figure 5 A longitudinal slice taken from each 3-d data set is shown. Data has been colour coded with an arbitrary scale to show the increase in signal intensity as bound water in the bud tissues is freed.

men by drilling out the base, so that the same stem can be imaged through the winter. At each imaging date, a series of 2D and unweighted 3D spin echo images are acquired using standard Bruker pulse sequences.

We have obtained 2D and 3D images showing the development of leaf primordia and flower initials with the bud (Fig. 5). From these images, T_1 (spin-lattice relaxation) and T_2 (spin-spin relaxation) times can be calculated to establish the concentrations of ^1H protons from the water within the bud. Dormancy in raspberries is a dynamic process, and the use of NMR micro-imaging of the water status in raspberry buds presents a powerful technique for direct, non-invasive observation of developmental and temporal changes of internal structures in raspberry buds.

To complement the NMR micro-imaging, optical imaging by CLSM is being used to observe the structural changes within buds as flower primordia initiate and differentiate. Excised raspberry buds are fixed using 5% glutaraldehyde in PIPES fixative. The buds are stained using Safranin-O (0.01%) and embedded in Araldite® resin[11]. A Bio-rad MRC 1000 CLSM is used to optically section the embedded raspberry buds.

CLSM optical sectioning is a simple and reliable method to visualise flower initiation and differentiation in raspberry buds. Clearly defined images at both the macro and micro level have been obtained (Fig. 6).

Crop productionThe removal of apical stem parts in the winter, or 'tipping', has been standard practice in the field cropping of raspberries in the UK for many years, for ease of management and improved yields[12]. Growing of protected raspberries using long-cane methods has also involved the use of tipping on canes entering the glasshouse from cold store (Raffle, Pers. Comm.), again for ease of management but also in an apparent attempt to reduce the effects of apical dominance as expressed in the uneven breaking of buds down the cane. Such unevenness is a major factor in reducing yields in protected raspberry cropping.

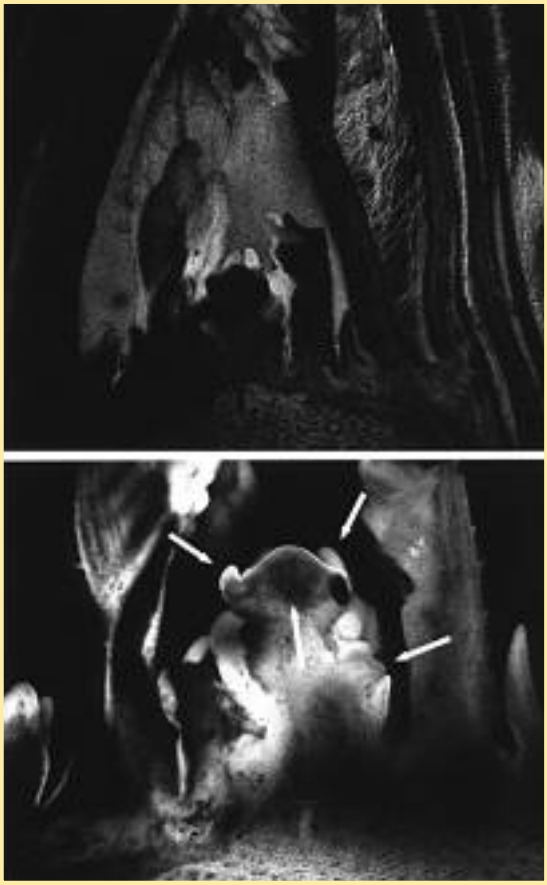

Figure 6 Text: CLSM optical section through a resin embedded bud stained with Safranin-O. a) x4 objective: the terminal flower primordia can be seen in the centre, with developing leaves and protective bud scales to the right and left. b) x10 objective: terminal flower with arrows pointing to the rounded torus and sepal rudiments, a secondary flower stem can be seen on the right of the terminal flower.

In a trial to examine the effect of tipping on long-canes under glasshouse production, canes of Glen Moy and Glen Clova were tipped at two heights, 1.5m and 1.8m, on entering the glasshouse. Bud development (assessed on a scale of 1-6, with 1 being unbroken and 6 fully opened), fruit yield and berry weight were compared to untipped controls.

The use of tipping is often thought to have a beneficial effect on the reduction of apical dominance and evenness of bud break, but our studies showed that there was generally little effect on the uniformity of bud break along the cane. Results of tipping on bud-

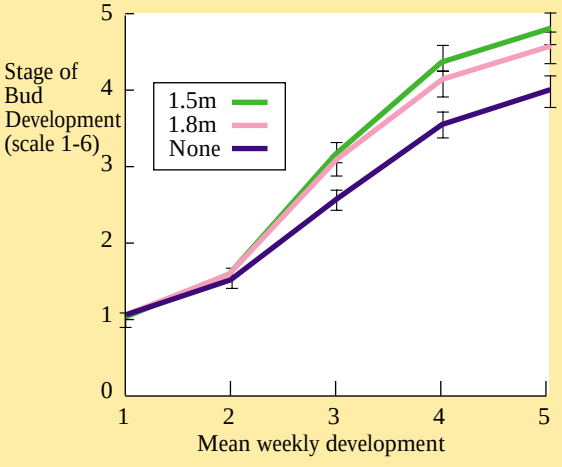

Figure 7 The effect of 'tipping' on the rate of bud development in Glen Moy.

break varied between varieties: there was an increase in the rate of bud development in Glen Moy (Fig. 7) together with larger fruit size. No comparable effects were observed in Glen Clova, with evenness of bud break assessed by the percentage of unbroken buds, similar in all three treatments (11.3%, 10.7% and 13.3 % for untipped, 1.5m and 1.8m respectively). Yields between the treatments were similar. The tipped canes with less buds per cane produced more fruit per lateral compared to the untipped canes in both cultivars.

One developing school of thought is that uniform bud break will occur in the cropping region of the cane if the cane has received adequate chilling. Uniformity of cane diameter and 'wood' maturity are thought to be important with regard to chilling requirement. If the cane is uniform in diameter, then the lower buds should require the same or only slightly more chilling than the upper buds. Taper at the base of the cane and woodiness of the lower stem may increase the chilling requirement of buds in the lower region of the cane.

Correct cold storage conditions are also vital for good bud break. If the cane is able to transpire during storage, the likelihood of bud dehydration is increased. Temperatures that reduce transpiration and respiration to a minimum are ideal. Infection of buds by fungal pathogens is also exacerbated if temperatures in the cold store are too high. Other workers have successfully stored canes at temperatures as low as -2°C, primarily to control infection by *Botrytis*[13]. Work at SCRI is currently in progress to examine these hypotheses and the interaction between cane maturity, chilling requirement and storage at sub-zero temperatures.

Future The ability to extend the fresh fruit season in the UK will confer benefits in terms of diversification, reduction of the considerable imports of fresh and processed fruit, and increase availability to consumers.

Through a greater understanding of the physiological processes involved, and quantification of the ideal conditions for long-cane production, maximum fruiting potential from this system can be achieved. At present, cultivars that were originally bred for field production are being used for out-of season production under protected cropping systems. Material from the extensive germplasm collection at SCRI is being trialled in the long-cane project to quantify the differing responses of cultivars to the long-cane system. New cultivars with useful traits that have been identified through the long-cane project, e.g. low chilling requirement, can then be selected specifically for protected cropping in the UK.

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Efficient genetic transformation of grain legumes for improved fungal resistance

Jane Miller, Geetha Shilvanth, B. Williamson & G. Ramsay

Grain legumes are one of the most important crop groups in world agriculture, improving soil fertility and yielding nutritious protein-rich seeds for human consumption or animal feed. The most widespread problems experienced by growers are diseases caused by fungal pathogens. *Botrytis* grey mould, for example, can devastate chickpea crops in the Northern part of the Indian sub-continent, risking the food security of resource-poor farmers. Even where fungicides are available and affordable, their use can render the growing of the crop less economic. In Europe, pea and beans are grown primarily on a farm scale for their protein-rich contribution to animal feed. However, home-grown grain legumes have to compete with soya meal, mostly imported from the United States. Only one third of Europe's requirement for protein-rich materials is met from within its borders, ultimately due to the cost of home-grown legumes relative to the current world trade price of soya meal. Better inherent fungal resistance in the grain legumes grown in Europe could help to improve this situation. In addition to economic factors, environmental benefits would also arise from a reduction in the use of fungicides on grain legumes.

If public concerns over the release of GM crops and the use of GM foods can be answered, new forms of fungal resistance in grain legumes provide one possible application of the technology with clear environmental benefits. Other developments of grain legumes are possible with GM technology: reduced anti-nutritional factors, improved amino acid balance for animal feed and insect resistance for example. The available genetic transformation techniques for grain legumes are, however, inefficient and unreliable. Two research projects now nearing completion were targeted at the improvement of transformation methods for grain legumes, and their use to introduce new genes giving enhanced resistance to fungal pathogens. Here we describe the techniques developed in these projects.

Progress in the genetic transformation of grain legumes has been hampered greatly by their generally poor response in tissue culture. Even in pea, where transformation has been reported by several groups through the 1990s, methods are frequently very inefficient and not readily repeated. Although capable of being transformed, faba beans and chickpeas have been even more difficult to transform than peas. A common approach to achieving an efficient method has been taken here in all three crops. Given the poor *de novo* regeneration response seen for these crops *in vitro*, a focus was made on methods using *Agrobacterium* inoculation of meristematic tissues. *Agrobacterium* naturally performs genetic engineering to introduce its own genes into plant cells when acting as the crown gall pathogen; this process is used with disarmed forms of the bacterium to introduce new genes under the control of the investigator. Seedling tissues with pre-existing meristems are ideal starting material for meristem-based transformation, being both readily available and able to grow to form a mass of shoot buds in culture. The target meristems need to be disrupted in some way to allow the *Agrobacterium* cells access to regenerable cells in the meristem. *Agrobacterium* inoculation of disrupted meristems is known to give inefficient gene transfer to plant cells with a resulting low frequency and unreliability of stable genetic transformation events. The approach taken here was to enhance the efficiency of the *Agrobacterium*-plant interaction by monitoring the expression of the GUS marker gene during the initial few days after inoculation. Various changes to the culture regime and to the inoc-

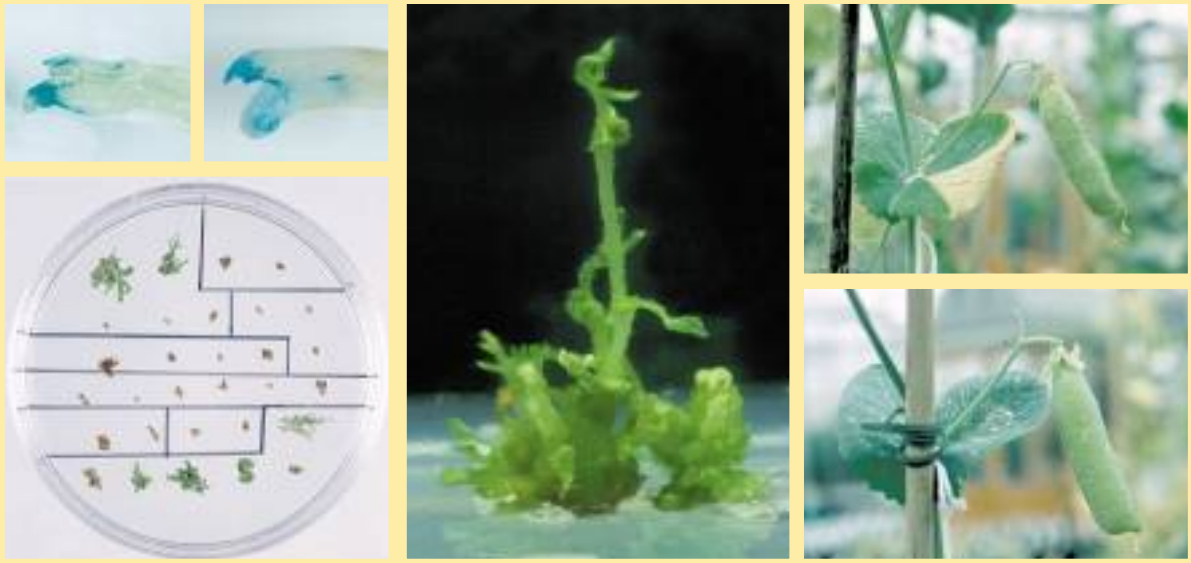

Figure 1 Transformation protocols developed for grain legumes: optimised transient expression of the GUS gene in chickpea shoot slices (upper left), selection of pea shoot clusters on medium with phosphinothricin (lower left), recovering chickpea shoot *in vitro* (centre), susceptible pea control PPT leaf paint test in pea for the *bar* gene (upper right), resistant pea leaf paint test showing expression of the *bar* gene (lower right).

ulation process have resulted in a dramatic improvement in the efficiency of this initial *Agrobacterium*-plant interaction. These, together with modifications to the subsequent tissue culture steps, combine to give efficient and repeatable transformation in both chickpeas and peas. Faba beans have also been successfully transformed by these methods.

The outline of the transformation system is given in Figure 1. Several steps in the process are important for the success of the method. The *Agrobacterium* strain/plasmid combination, the plant genotype and the kind of explant influence the outcome. The conditions of the *Agrobacterium* culture, and the plant explants exposed to it, have a large influence on the amount of GUS gene expression seen. Optimal conditions with the AGL1 strain and the pGIN1 plasmid include the use of an *Agrobacterium* culture at the end of the logarithmic growth phase, and an induction period in a low pH medium with a modified composition. Crucial to the high levels of GUS expression seen in the explants is a careful drying of the explants after being submerged in *Agrobacterium* suspension, and subsequent culture on filter paper rather than in full contact with the culture medium. All of these factors contribute to reliably high levels of GUS expression in the explants a few days after inoculation, as detected in histochemical assays (Fig. 1) and quantitative enzyme assays (Fig. 2). When applied to experiments with subsequent extensive shoot proliferation on thidiazuron-containing medium, followed by selection on phosphinothricin-containing selective medium, transgenic shoots were produced at a high efficiency. In one experiment, eight verified independently transformed shoot clones were produced from only 50 seeds inoculated. The long-term average is lower than this, but in 16 experiments in chickpea, 6% of seeds

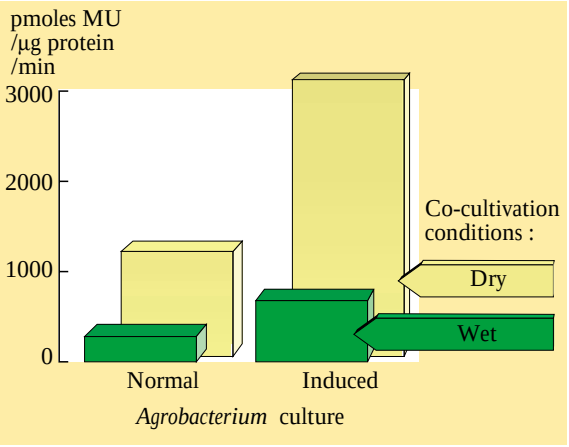

Figure 2 The effect of improvements in transformation protocols on the levels of transient expression of the GUS gene in chickpea seedling shoot slices.

overall gave rise to transformed shoot clones. Such frequencies of transformation are unprecedented, and indicate that pea and chickpea transformation is now both routine and efficient. Faba beans, previously very hard to transform, also have been transformed with this method.

The way is now open to the testing of approaches designed to enhance fungal resistance in grain legumes. Most of these approaches use genes from other food crops. Polygalacturonase-inhibiting protein (PGIP) has the ability to inhibit one of the key enzymes used by *Botrytis* and some other pathogenic fungi during invasion, and may therefore delay the pathogen long enough for other defences to take over. PGIPs from raspberry and kiwifruit are being used in this work. Phytoalexins, secondary compounds made by many plants, have a direct inhibitory effect on the growth of fungal pathogens. Stilbene synthase uses common precursors to generate the phytoalexin resveratrol, regarded as a desirable component of red wine. This enzyme is present in groundnuts but not in the grain legumes under consideration here. The pea pathogen *Mycosphaerella pinodes* is known to have evolved specific defences against pea phytoalexins; it can be expected that such specialist pathogens may not have specific mechanisms for coping with stilbene phytoalexins. Further natural defences used by plants are targeted at lysing fungal cell walls. Glucanases and chitinases are components of normal pre-existing defence mechanisms in many plants and function in this way. The endochitinase derived from the biocontrol fungus *Trichoderma harzianum* appears to be particularly effective in inducing resistance to fungi and this version of endochitinase is being tested here. All of these genes are being used in a nine-partner European project to investigate their potential for the replacement of fungicide applications in grain legumes. The possibility of producing chickpeas resistant to *Botrytis* grey mould is also being investigated in a DfID-funded collaboration with the International Crops Research Institute for the Semi-Arid Tropics in India. Although such approaches hold much promise for more effective and environmentally-friendly means of controlling fungal pathogens, this work is still clearly at the exploratory stage, requiring further development and testing before application in the field.

Acknowledgements

We wish to acknowledge funding from the European Commission's FAIR programme, and the Department for International Development.

New Blackcurrant cultivars

Ben Gairn (F4/1/67) is derived from a cross between Ben Alder and the Russian cultivar Golubka, and provides good yields of early-ripening fruit. The main agronomic advantage of this cultivar is its resistance to blackcurrant reversion-associated virus (BRAV), derived from Golubka.

The growth habit is fairly compact, and flowering is early (c. 7 days before Ben Lomond), with consequently a small risk of frost damage in some years, although performance to date has not shown undue losses.

Fruit quality is good in terms of juice colour, although ascorbic acid levels are fairly low, and the fruit is acceptable for commercial juice production. Berry size is fairly large, and harvest is usually c. 8-10 days before Ben Lomond.

Resistance to foliar diseases is good.

Ben Hope (C1/9/10) is derived from a complex cross between Westra x (238/36 x EM21/15). It is a very vigorous and upright-growing bush, and most importantly has a high level of resistance to blackcurrant gall mite based on the *Ce* gene. As a consequence, it may be amenable to low-input growing systems.

It flowers c. 2 days after Ben Lomond, but its harvest date is usually up to 10 days later than Lomond. Yields are consistently high, with medium-sized berries, and the fruit is acceptable for commercial juicing on all grounds, including good sensory quality.

Ben Hope has good resistance to both mildew and leaf spot.

The plant variety rights for both cultivars are held by SmithKline Beecham plc.

New Potato cultivars

Amour

Amour is a multipurpose, early maincrop cultivar, which was placed on the National List in 1998. It has proved very successful in overseas trials, particularly around the Mediterranean basin in such countries as Cyprus, Egypt, Morocco and Israel. It is the product of an SCRI parent crossed with Cara, and produces a bold sample of large, red-eyed, oval tubers. Earlier than Cara, it has inherited its parent's resistance to the golden potato cyst nematode (*Globodera rostochiensis*, Ro1 pathotype). It is marketed through the Zenith group.

	Amour	**Blush**
Origin :	9559AB2 x Cara	15005A2 x 12380AC2
Year of cross :	1982	1987
Maturity	Early maincrop	First Early
Wart	Susc.	Field immune
Late blight (f)	5	1
Late blight (t)	5	4
Gangrene	5	4
Common scab	4	7
Virus:		
PVY	2	8
PLRV	4	6
Spraing	-	4
PCN:		
G. rost.	9	Partial R
G. pall.	Susc.	Partial R
Blackleg	-	5

Blush

Blush is a first early cultivar, which was placed on the National List in 1998. It produces high yields at both early and mature lifts and has attractive oval cream tubers with pink eyes and white flesh. Blush has good levels of resistance to virus Y and Common scab, and also partial resistance to both the golden *(G. rostochiensis)* and white *(G. pallida)* potato cyst nematodes. It is marketed through the Agrico / Greenvale AP consortium.

Plant biochemistry and phytochemistry

Howard V. Davies & William W. Christie

The furore over genetically modified foods has accelerated since the turn of the year and must have an impact in the short- to medium-term on all concerned with research and development in plant biotechnology and related sectors. The public should be made aware of the fact that molecular sciences now provide the indispensable tools with which to dissect plant form and function, tools which in the hands of professionals add considerably to the basic knowledge required for crop improvement at many levels. Part of the GM debate centres around potential impacts on the environment, and on the chemistry and composition (safety) of the end products, whether destined for human or animal consumption. The SCRI's skill base facilitates high quality research in all of the above areas, as exemplified by activities in the biochemistry and phytochemistry programmes. This applies to plants whether genetically modified or not. The following overview gives examples of progress in some key areas over the past year.

Carbohydrate research Sink-source relationships play an important role in regulating carbon partitioning to commercially important storage organs such as potato tubers. When developing tubers are detached from the mother plant, there is a significant reduction in the starch synthetic capacity of the tubers, accompanied by a rapid reduction in ADP glucose and an increase in hexose phosphates. The results indicate that one of the first responses to tuber detachment is a reduction in carbon (or ATP) import into the amyloplast. It is speculated that the rate of metabolic exchange between the cytosol and the amyloplast in storage parenchyma cells of developing tubers, depends upon a continuum between the storage organ and the photosynthetic machinery. However, results to date indicate that sucrose is not directly involved in

this process. Also related to sink-source interactions, a programme on the metabolism of tubers during dormancy break (EU-funded) has shown that initial bud growth is sustained with 'soluble' carbon reserves present in the tuber throughout the dormancy period. It is speculated that activation of transport mechanisms and channels from the tuber storage parenchyma to the buds, is a key process in dormancy break.

A transgenic approach is exploited to investigate the role of a range of target genes on carbohydrate metabolism in potato. For example, two α-glucosidases have been cloned, and the effects of down-regulating these genes, using an antisense approach, is well underway. Alkaline invertases from potato and sugar beet have also been cloned and an investigation of the function of these genes is in progress. Maize genes involved in the biosynthesis of starch have also been expressed in potato, and the effects on starch determined. Genes from micro-organisms are also under evaluation with a view to generating novel starch types. A SOAEFD Flexible Funded programme on the processing potential of starches from Scottish-grown crops has been completed and the report submitted. One of the outcomes of the programme is a database containing information on the composition and properties of 619 samples of starch. While all analyses were not carried out on all samples, it is probably the largest single database available on starch composition and properties. The report allows a comparison of the properties and composition of many industrially-produced cereal and potato starches with those produced in the lab from equivalent plant sources. In addition, the scale of the material collected allows many comparisons of the influence of genotype, as well as of genotype x environment interactions, to be derived. The programme also dealt with the relationships between physical and chemical composition of starch, with a view to predicting more complex behaviour from simpler analyses. Considerable use was made of statistical and neural network analyses.

Mathematical biology and biochemical systems Theoretical work aimed at understanding the regulation, dynamics and thermodynamic properties of biochemical systems has progressed, with a major focus on the ability of these systems to adapt to different types of applied stresses. Hierarchical levels of organisation exist in living systems and a full understanding of biochemical functioning must account for the interactions within and between these different levels, and with heterogeneous and stochastically fluctuating environments. A central challenge is to understand how the complex reactions taking place inside cells remain co-ordinated under different environmental conditions and how they respond to changing conditions. Although biochemical systems are generally complex, all have two main features. One is that the activity of any enzyme species becomes saturated at some concentration, irrespective of any additional regulation. The other is that the changes in flux catalysed by any enzyme are not simply proportional to changes in substrate concentration, i.e. enzymatic kinetics are always non-linear. A linear simplification would require very restrictive conditions, which cannot generally be validated for biochemical systems. It is the interactions of these two main features that determine whether or not biochemical systems can adapt to different environmental conditions. Non-linearity may result in different types of temporal and spatial organisation in biochemical systems, and the simple fact that enzyme activity saturates, imposes conditions on these states in order for the reactions to remain co-ordinated. If these conditions are not satisfied, then the cell cannot function. Importantly, the conditions which are suitable for the co-ordination of biochemical systems under different environmental conditions can be determined, and collaborative research in this area is progressing.

A new collaborative project has been established with Strathclyde University and Glasgow University, with support from the BBSRC and EPSERC. A postdoctoral research assistant has been employed to work on the effects of environmental fluctuations on biochemical systems. In addition, collaborative research with University of Laguna, Spain, is underway with the aim of improving the efficiency of citric acid production in *Aspergillus niger* metabolism.

Free radical mechanisms and natural antioxidants in plants An EU-FAIR project has been initiated, involving collaboration between four SCRI departments and seven European and Israeli partners. The programme is investigating the oxidative processes associated with necrotrophic infection. SCRI is utilising both spectroscopy (EPR) and comparative biochemistry to study the complex events associated with pathogenesis. EPR spectroscopy has been used to study apparent shifts in redox status of tissues post-inoculation, whilst characteristic markers of lipid peroxidation have been quantified and correlated with the aggressiveness of the pathogen. EPR spectroscopy also has been used to investigate the effects of abiotic and biotic stresses on free radical processes in cereal

plants. The main applications this year have been on wheat and rice in collaboration with the Austrian Research Centre, Seibersdorf, and the International Rice Research Institute. The work with wheat has concentrated on the detection of free radical damage at relatively low levels of exposure to ozone. The EPR data indicate that there is a threshold exposure level, below which there is no effect on the free radical signal and above which there are major increases in free radical signal with relatively small increases in ozone concentration.

Experiments to identify free radicals generated in plant tissues as a result of physical damage, have been performed using the new spin trap DEPMPO, which has the advantage over its older analogue DMPO in that it is able to discriminate between $O_2^{\cdot -}$ and HO· adducts. Preliminary results indicate that HO· and C-centred radicals are the main products formed in a range of different tissue types. However, the relative intensities of the EPR signals from the two adducts are strongly dependent on the spin trap/tissue ratios and considerably more work is needed in order to understand the relevant reactions involved.

Extending our interests in the nutraceutical status of foods and the role of antioxidants, funding has been gained for work on the potential health-giving properties of the constituents of fruits such as strawberry, raspberry and blackcurrant. The work, funded by a SOAEFD Flexible Fund grant, involves collaborations with the Rowett Research Institute and Glasgow University. Individual antioxidants and natural synergistic combinations of antioxidant molecules will be identified and those with the most potent activities will be assessed in animal models for bio-availability, selective accumulation in tissues, *in vivo* antioxidant properties and their effects on biomarkers of disease risk. The results will provide the initial information necessary to genetically improve the antioxidant capacity of soft fruits, the ultimate aim being to provide an indigenous additional food source for improving antioxidant status *in vivo* - a prerequisite in establishing requirements for optimum health.

Fruit research In strawberry, a wide range of ripening-related genes has been isolated using cDNA AFLP-based approaches. Candidates genes have been selected for inclusion in a transgenic programme with the objective of improving a range of quality traits. As part of a SOAEFD Flexible Funded grant, protocols for two-dimension gel electrophoresis of strawberry proteins have been developed and exploited to sequence proteins which change in relative abundance during ripening. This proteomic-based approach has proved extremely complementary to the programmes on differential gene expression, identifying different targets for crop improvement. In parallel with molecular approaches, the use of modified atmosphere packaging to extend shelf life has been studied. Strawberries have been packaged in sealed plastic punnets using impermeable polypropylene film and three gas mixtures; 20% O_2:80% N_2 (air), 5 % O_2:5% CO_2:90% N_2 and 80% O_2:20% N_2. All gas mixtures dramatically reduced weight loss during storage, with air marginally the best. Maintenance of strawberry juice antioxidant status over relatively longer-term storage was achieved most effectively with high O_2 as the packaging gas. Overall, the fruit stored in the high O_2 mixture proved most successful with respect to maintained firmness, minimised cell wall breakdown and visual appearance. A more expansive article on the fruit packaging research is provided later.

NMR images of fruit infected with fungal pathogens such as *Botrytis cinerea* frequently show high intensity regions corresponding to the fungal lesion. To demonstrate whether or not such contrast is due to the fungal hyphae or to damaged host tissue, blocks of agar gel have been inoculated with spores and imaged by NMR. In the absence of any cell structure to be disrupted by the pathogen, no contrast was visible by NMR in the region infiltrated by the fungus. Thus, the dramatic NMR images of *Botrytis*-infected fruits are due to damaged host tissues. The non-invasive nature of NMR imaging has also proved useful in monitoring the water status of raspberry canes and buds to reveal large changes immediately prior to bud break. The subsequent fruiting of the bud monitored *in situ* is testament to the non-destructive nature of the technique.

Lipid research A new method for the determination of residual solvents in synthetic ^{13}C-labelled triacylglycerols has been developed using thermal desorption. Residual solvents such as acetone, acetonitrile and branched-chain hexanes can be quantified to 0.1 parts per million level. With this information, further purification steps could be performed to reduce solvent residues to levels acceptable for nutritional studies.

Work has continued on developing methods for structural characterisation of plant membrane phospholipids and glycolipids. A novel approach for determining molecular species composition, using liq-

uid chromatography (LC)-atmospheric pressure chemical-ionisation mass spectrometry of diacylglycerol nicotinates, has been developed further to cover a wider range of lipids, and the LC separation has been optimised. A two-dimensional thin-layer chromatographic procedure has also been developed to separate individual phospholipids and glycolipids and has been applied to leaf tissue of *Arabidopsis* wild-type and mutants with altered lipid compositions. The method will be used to isolate individual lipids for subsequent quantification, and fatty acid and molecular species determination.

A novel fatty acid (octadeca-8,10-dien-12-ynoic acid) has been identified in the seed oil of *Tanacetum corymbosum* by chromatographic, spectroscopic and degradative procedures. 4,4-dimethyloxazoline derivatives are widely used for GC-MS analysis of fatty acids, but applications to natural fluorinated fatty acids and synthetic fatty acids, labelled with stable isotopes, have shown that rearrangements can occur in the mass spectrometer that can confuse the interpretation of results.

Compound-specific isotopic analysis of fatty acid methyl esters has yielded promising results, particularly for applications to human lipid metabolism. On-line pyrolysis of methyl esters, separated by GC, to hydrogen, followed by continuous flow measurement of the deuterium/hydrogen ratio, gives reliable results when there is a near natural abundance distribution of deuterium-containing isotopomers. This provides a convenient and sensitive method for measuring deuterium incorporation from labelled body water, and hence fatty acid synthesis *de novo*. With partners in an EU project, the processes of desaturation and chain extension are being studied using uniformly ^{13}C-labelled linoleic acid and following the incorporation into arachidonic acid using GC-combustion-IRMS to detect the low enrichment.

Plant volatiles A range of methods has been evaluated for identifying volatile compounds released by, or present, on the surface of blackcurrant leaves. Analysis of the leaf surface extracts revealed that the main constituents were mono- and sesquiterpene hydrocarbons. Similar results were obtained using solvent elution and thermally desorbed polymer-entrainment, but 'green leaf' volatiles and a homoterpene were also detected. Thermal desorption resulted in the production of at least two artefacts due to heat- or metal-induced rearrangements. Steam distillation was the least satisfactory method, causing rearrangement and oxidation of some terpene constituents. The optimum method for characterising plant-derived odour plumes proved to be solvent elution combined with polymer-entrainment.

As part of an investigation into the ecological chemistry of insect predator/prey interactions, the surface and internal lipids have been analysed from two species of ladybird and their principal food source, the pea aphid. The beetle lipid consisted of hydrocarbons, fatty acids, alcohols and defensive alkaloids (external and internal) and triacylglycerols (internal). Aphid lipid (internal) consisted almost entirely of triacylglycerols, but with shorter acids (C_6, C_{12}, C_{14}) on the glycerol backbone than was found (C_{18}, C_{20}) in the equivalent ladybird compounds. The nature of the ladybirds' internal lipid depends on the type of food source. Insects fed on aphids showed evidence of incorporation of fatty acids derived from aphid triacylglycerols into their own triacylglycerols.

In a collaborative study with the Department of Biological Sciences, Stirling University, the floral volatiles contributing to the aroma of the cut flowers of three varieties of the Sweet Pea (*Lathyrus odoratus*) have been characterised. Major components included (E)-b-ocimene, linalool, nerol, geraniol and α-bergamotene.

Plant fibres research An EU-funded programme on Reed Canary Grass, as a source of pulp for paper and biomass, has been extended due to commercial interest. The plots at SCRI continue to thrive and produce high yields of biomass. However, in common with all the more southerly partners, the lack of a really cold winter with a clear cessation of growth has hindered this programme. The quality of the material produced in all countries, except Sweden and Finland, may be inferior due to too high levels of protein. The group at SCRI has the responsibility for quality assessment over the whole programme in relation to cell wall composition and use of IR spectroscopy to predict quality.

A new initiative is underway into the use of hemp fibre, both untreated and treated, as a source of strength to recycled paper. An inclusion level of 1-2% is likely to be feasible compared with 20% for other fibre sources.

The use of trifluoroacetic acid in the characterisation of plant cell walls is continuing. A fragmentation scheme has been proposed and the fragments are currently being assessed to show how some of the cell

wall macromolecules are connected to build up the three-dimensional network. Improvements in the scheme also are being investigated, with a major advance being the elimination of water during fractionation and hence reducing the opportunities for acid-catalysed hydrolysis of susceptible chemical bonds. Based on these experiments, a simple procedure for the determination of cellulose and total non-cellulosic polysaccharides in cell wall samples is being proposed. Oxone bleaching/delignification studies have now been completed on Jute and Sisal. Oxone proved to be effective at removing lignin and bleaching at low temperature (70°C) and low doses (5% Oxone) without any deleterious effects on the fibre performance. Studies with TAED, a novel environmentally benign delignification/bleaching agent, have been initiated using cereal straw as the sources of fibre. Preliminary results suggest that it is equally effective, if not better than, conventional chlorine-based bleaching strategies.

An initial study into the replacement of PVC/polyester fibres systems with coated plant fibres has been completed and several conclusions drawn. For example, polyurethane coatings are amenable for use with woven cellulosic materials and are capable of inhibiting fungal degradation of matrix flax fibres. However, co-blending of specific high strength synthetic/natural fibres would be required to increase tensile strength to values similar to high strength PVC fabrics.

With respect to the biochemistry of cell walls and lignification processes, oxidase activity has been shown to be ubiquitous in extracts from lignifying, developing xylem tissue and from a taxonomically-diverse range of tree species. This suggests that these enzymes are required for lignification reactions. In addition, the enzymes from angiosperms and gymnosperms have specificities for monolignol oxidation that mirror the monolignol composition of the lignins from these species. A quasi-proteomic approach has been used to identify proteins that are differentially expressed in the developing xylem of compression and normal wood of conifers. A number of candidate cell-wall associated proteins have been identified that may be responsible for the altered cell wall structure and composition characteristic of compression wood. In particular, one compression-wood specific protein yielded an amino-terminal protein sequence that was homologous with known plant laccase genes. This represents the first laccase to be identified from conifers.

Work in research related to the use of stable isotopes is presented in a separate research article.

Effect of modified atmosphere packaging (MAP) on soft fruit quality

D. Stewart, J. Oparka, C. Johnstone, P.P.M. Iannetta & H.V. Davies

Soft fruits such as strawberries and raspberries, have a very short post-harvest shelf-life which is exacerbated by infection with grey mould caused by the fungus *Botrytis cinerea* (Fig. 1). There are significant commercial gains to be made by minimising such deteriorative losses in both retail outlets and in the home. Strategies to extend the shelf-life of fruit include traditional breeding, genetic manipulation and post-harvest treatment of the fruit or its storage environment. Thus far, traditional breeding of strawberry and raspberry has had only limited success in extending shelf-life. Genetic engineering is an attractive prospect as the concept is already proven for tomato and melon, where restricting ethylene production improves fruit longevity in store. As yet, such technology is unproven for raspberries and strawberries. Although ethylene is also likely to play a key role in regulating raspberry quality traits[1], mechanisms controlling strawberry post-harvest quality are less obvious. Whilst the biological processes underpinning such processes are being elucidated, other options are available to the commercial sector. These include post-harvest treatments such as coating the fruit with a protective film e.g. chitosan, or storage of fruit under a controlled or modified atmosphere to minimise deteriorative losses.

Figure 1 Strawberry infected with *Botrytis cinerea*.

In retail outlets, strawberries and raspberries are generally stored in closed, perforated-top containers. Within these containers, the fruits continue to respire the 'trapped' air until the CO_2 concentration rapidly approaches the critical 10-15% level necessary to inhibit *Botrytis* growth. However, depending on the state of the fruit at harvest, the time taken to pack and the storage temperature, *Botrytis* infection may be significant before the CO_2 concentration has reached the critical level required to suppress fungal growth. An alternative approach is to flush the package with known gas mixtures to provide, directly, a modified storage atmosphere and/or to use impermeable or selectively permeable packaging films to maintain the best environment for as long as possible. The approach generally involves gas mixes confined to 5-15% CO_2, 2-5% O_2, with N_2 as the remainder. The elevated CO_2 concentration at packaging generally rises during the first 12 h and inhibits *Botrytis* growth. However, one drawback is that the continued presence of an elevated CO_2 environment induces a concomitant decrease in the pH of the fruit, leading to a deleterious 'fizzy' or sharp taste. This can be alleviated by employing one of two methods: (1) A novel modified atmosphere packaging (MAP) gas system with O_2 as the dominant gas; and (2) A semi-permeable membrane which facilitates the diffusion of moisture and gases establishing an equilibrium state which, once optimised, retards deterioration of the fruit. At the SCRI, these strategies have been assessed for the possibility of extending storage life of both

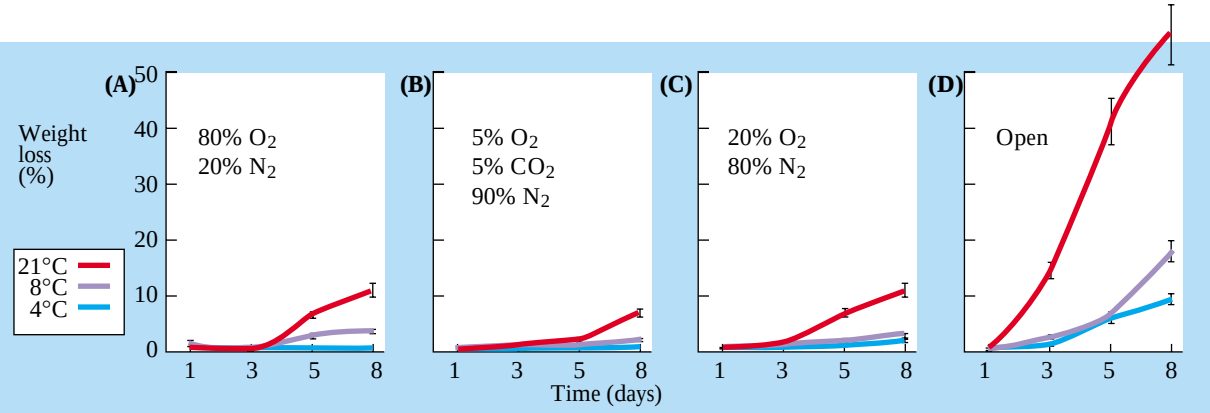

Figure 2 Weight loss of strawberries packaged in (A) 80% O_2:20% N_2; (B) 5% O_2:5% CO_2:90% N_2; (C) 20% O_2:80% N_2; and (D) in the absence of packaging.

strawberries (high O_2 approach) and raspberries (semi-permeable membrane approach).

Effect of elevated CO_2 or O_2 on strawberry quality Strawberries have previously been the subject of MAP studies largely relying on the use of elevated CO_2 and reduced O_2 levels[2]. These have met with limited success, the main problem being the progressive increase in CO_2 concentration leading to increased acidity in the fruit. The advent of MAP approaches using elevated O_2, rather than elevated CO_2, appears to alleviate this problem with the added advantage of retaining fruit firmness. This system has been used successfully with meat and, more recently, with cut vegetables[3]. The use of O_2 at concentrations much greater than that present in air (50% as compared with 20%), prevents microbial spoilage by dramatically reducing the activity and proliferation of lower organisms. Its use, therefore, should help inhibit the growth of microbes unaffected by ≤20% CO_2[4]. Little is known about the biochemical changes induced in fruit by modified atmospheres and, particularly, the effects of elevated O_2. Here we report the results of an investigation on the impact of MAP on cell wall hydrolase enzymes, which may have a role in regulating fruit firmness, and antioxidant status, an increasingly important parameter which gauges the potential of the fruit to quench free radicals.

Experimental approach Fruit of cultivar Elsanta were picked from field-grown crops at the red-ripe stage, stored at 4°C for 1 h and packaged in containers held in impermeable polypropylene bags (OPP1 film). Packaging was carried out with an industrial packager (CVP Systems). The gas mixtures used were: (a) 20%O_2:80% N_2 (air-control), (b) 80% O_2:0% N_2 (elevated O_2) and (c) 5 % O_2:5% CO_2:90% N_2 (elevated CO_2). The packaging procedure involved two cycles of ambient gas removal/packaging-gas flushing prior to sealing of the packaging film. The packaged fruit were stored at 4°C, 8°C and 21°C and sampled at set periods over 11 days. Non-packaged fruit were also stored and sampled for comparative purposes.

Effects on quality parameters There was little to distinguish between the different packaging gases with respect to fruit weight loss. All gas mixtures were more effective at loss reduction than open packaging (Fig. 2). Not surprisingly, weight loss was inhibited at the lower storage temperatures. An examination of the concentrations of gases within the packaging revealed an increase in CO_2 (to a threshold of *c.* 50-60%) and a decrease in O_2 over the 11 day storage period, irrespective of the original gas mixture used (Fig. 3a-c). However, within the important initial stages of storage, CO_2 reached the level known to inhibit *Botrytis* infection (>10% CO_2) when fruit was stored under elevated CO_2. This was true for storage at 4°C and 8°C. Not surprisingly storage at 21°C rapidly generated CO_2 concentrations which impaired flavour by raising fruit acidity. Storage under air at 4°C and 8°C resulted in a gradual production of CO_2 but the inhibitory CO_2 level for *Botrytis* was only reached after 4-5 days, too late to be effective as a storage regime. Fruit packaged under elevated O_2 experienced a steady reduction in O_2 levels at 4°C and 8°C but these remained above the important 50% concentration over the first 4 days of storage, thereby inhibiting microbial growth[3].

Activities of cell wall hydrolysing enzymes The enzymes β-Galactosidase (β-Gal), arabinofuranosidase (Arab) and cellulase (Cx) have previously been identified as important in the soft fruit ripening processes[5], with β-Gal and Arab thought to be responsible for trimming the constitutive arabinogalactan-substituted

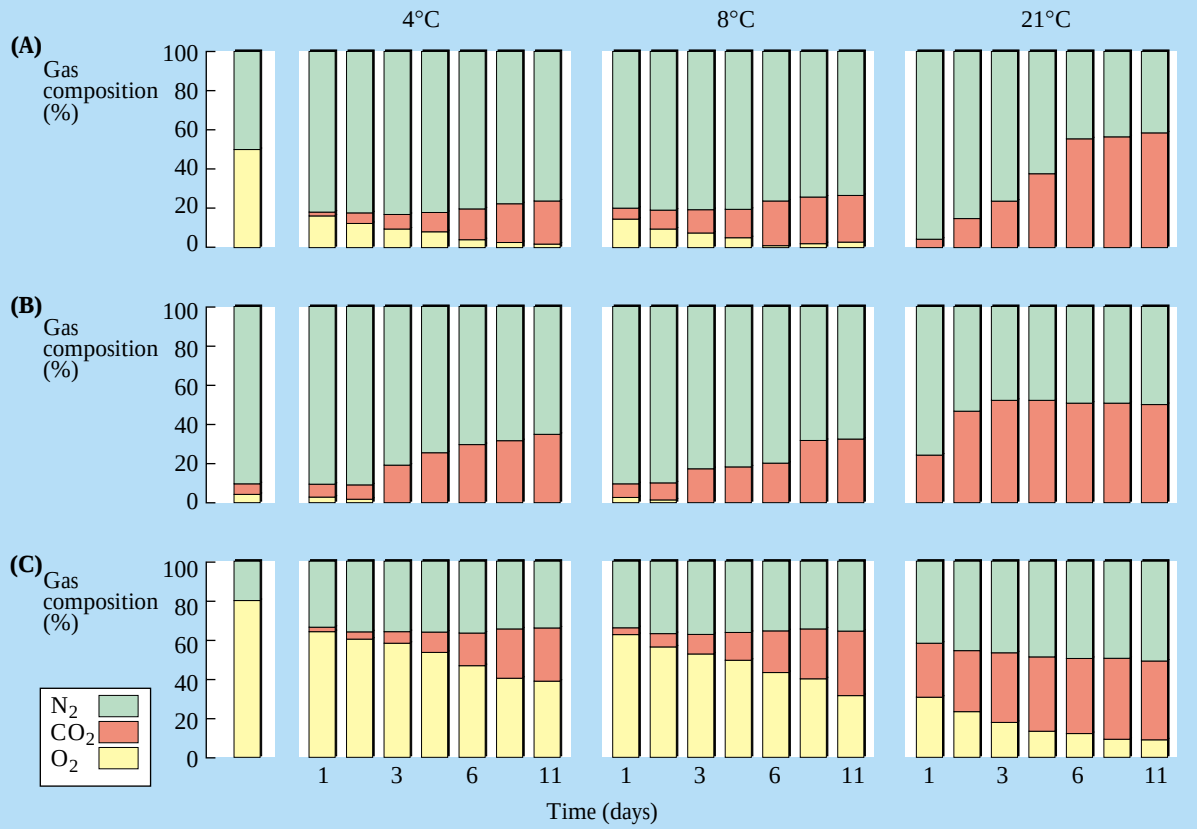

Figure 3 Gas composition of strawberries packaged in (A) 20% O_2:80% N_2; (B) 5% O_2:5% CO_2:90% N_2; and (C) 80% O_2:20% N_2. (C - control cylinder gas. Time t = 0).

rhamnogalacturonic acids prior to extensive pectin solubilisation. There was little difference in β-Gal and Arab activities between the gaseous storage regimes during the first 5 days of storage at 4°C and 8°C. However, enzyme activities were elevated during storage at 21°C (data not shown).

Cx activities varied with storage temperature and gaseous environment (Fig. 4). In all cases, activities were greatest in fruit stored at 21°C. Both the air- and elevated CO_2-packaged fruit showed progressive increases in Cx activity from 4°C to 8°C, more so under air. However, elevated O_2-packaged fruit showed no difference in Cx between the two low-storage temperatures but activity was again elevated at 21°C. In fact, there is no significant change in Cx activity throughout the period of storage at 4°C or

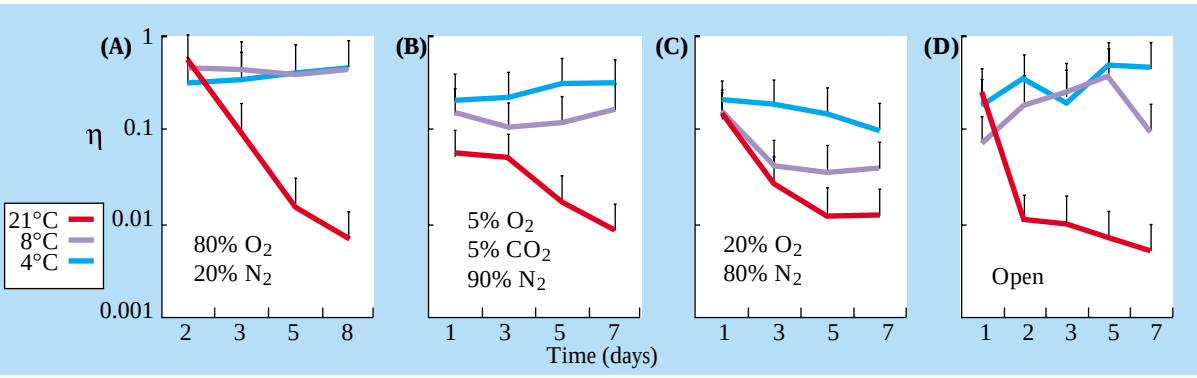

Figure 4 The effect of packaging gas on cellulase activity. NB. Cellulase activity is inversely proportional to viscosity in this assay.

Time (days)	Air 4.0	Air 8.0	Air 21.0	Elevated CO_2 4.0	Elevated CO_2 8.0	Elevated CO_2 21.0	Elevated O_2 4.0	Elevated O_2 8.0	Elevated O_2 21.0
1.0	*****	*****	***	*****	*****	***	*****	*****	***
2.0	*****	*****	*	*****	*****	*	*****	*****	**
3.0	*****	****	*	****	****	*	*****	*****	**/*
4.0	*****	****	*	****	***	*	*****	*****	*
6.0	****	****	n.a.	***	**	n.a.	*****	****	*
8.0	****	***	n.a.	***	**	n.a.	*****	***	n.a.
11.0	***	**	n.a.	**	**	n.a.	****	*	n.a.
30.0	***/**	***/**	n.a.	*	*	n.a.	****	*	n.a.

***** ➔ * : Decreasing numbers of stars indicates a decreasing degree of firmness. n.a. - this fruit had essentially disintegrated and was unsuitable for testing.

Table 1 Effect of MAP regimes on subjectively scored strawberry fruit firmness.

8°C under elevated O_2. This is important since the expression of Cx genes increases substantially in ripening strawberry fruit (Giorgio Casadoro, pers. comm.). Subjective firmness measurements of the packaged fruit indicated that, at all temperatures and sampling periods, the fruit stored in high O_2 were the firmest (Table 1).

Fruit antioxidant status A parameter becoming increasingly important with respect to fruit and vegetables is antioxidant status - the ability to inhibit the formation of free radicals. These highly reactive moieties have been repeatedly linked with many degenerative diseases in humans. The use of elevated CO_2 as the packaging gas reduced the overall antioxidative capacity, in particular during the important initial storage period (Fig. 5). The antioxidant status of air-packaged fruit initially decreased but subsequently increased. This may be due to microbial fermentation reactions producing acetaldehyde which would artificially raise antioxidant status. Fruit stored under elevated O_2 exhibited good antioxidative capacity over the first 4 days of storage but this declined with prolonged storage, possibly due to O_2-promoted oxidation of the constitutive anthocyanins and phenolics. There was visible evidence of mild bleaching in skin colour accompanying prolonged storage (data not shown). However, over the all-important first four days of storage the effect of elevated O_2 on antioxidative status was minimal.

The impact of elevated O_2 packaging on fruit longevity is clearly seen in Figure 6 (unpackaged fruit are shown for comparison). Strawberries were stored for 60 days following packaging under high O_2. On opening, the fruit were definitely edible, whilst lacking the 'tartness' associated with fresh fruit. This is clearly an extreme exercise but demonstrates the efficacy of elevated O_2 storage.

Modified atmosphere packaging of raspberry fruit A major advancement within the MAP industry has been the development of P-Plus™ packaging film. This film allows the diffusion of moisture and has a gaseous permeability that can be varied to suit the high rates of moisture loss and respiration that are

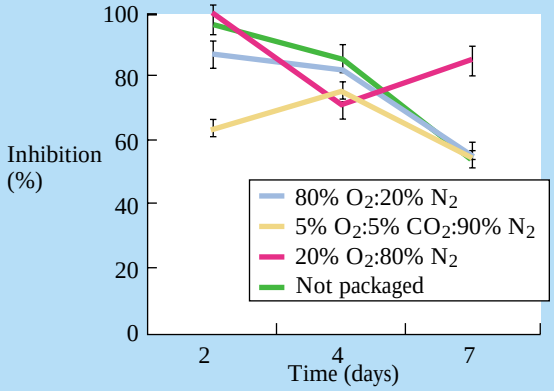

Figure 5 The effect of packaging gas on strawberry antioxidant status. Both packaged and non-packaged fruit were stored at 8°C

Figure 6 Strawberry packaged in 80% O_2: 20% N_2 after 60 days and a control open container.

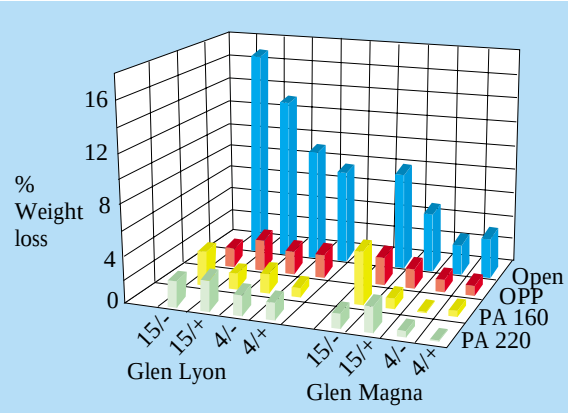

Figure 7 Effect of modified atmosphere packaging (no film (open), OPP, PA160, PA220), temperature (15 or 4°C) and ± Ethysorb™ on the weight loss of ripe raspberry fruits

characteristic of harvested raspberry fruit. The impact of packaging two varieties of raspberries, Glen Lyon and Glen Magna, with semi-permeable films on quality and shelf-life has been determined together with the effects of Ethysorb™, an ethylene 'scrubber'. Ethylene has been implicated as an important factor in determining post-harvest quality in raspberry fruit[5].

Experimental approach Freshly picked fruit were placed in plastic containers which were either left uncovered or sealed with impermeable (OPP) or semi-permeable films with high (PA220) and low (PA160) gas permeability. The fruit was stored at either 4°C or 15°C. Half of the fruit packages included an Ethysorb™ sachet. Shelf-life was defined as that period of time between harvest and the point when the fruit was no longer marketable either due to the development of off-odours, off-flavours or unacceptable appearance. Fruit was scored for incidence of *Botrytis* and changes in taste, colour, firmness and weight loss.

Effects on flavour and colour Packaging made no significant difference to the taste scores. Colour was maintained best at 4°C with the packaged fruit scoring higher than those left unpackaged (data not shown).

Firmness and weight loss The presence of Ethysorb™ helped retard softening during storage, particularly at 4°C. This supports our previous findings[5]. The combined benefits of Ethysorb™ and low storage temperature on firmness were most apparent for fruit stored using the less permeable OPP and PA160 packaging films. It is possible that the moisture retention abilities of these films contributed to prolonged fruit firmness. The most marked advantage of using packaging films was to restrict weight loss (Fig. 7), particularly at the higher storage temperature.

Resistance to *Botrytis* Overall packaging reduced the incidence of *Botrytis* infection compared to the unpackaged fruit.

If any one of the above quality parameters fell below a specified level the fruit would not be acceptable for sale. It was therefore possible to classify this data according to a consumer 'predicted acceptability rating' (PAR). The criteria for classification are defined in Table 2 where 'above-PAR' and 'PAR' represent good- or acceptable-quality produce, respectively. A rating of 'below-PAR' would not be marketable. Table 3 classifies each treatment according to those attributes that were 'below-PAR' and identifies the successful packaging treatments. It is evident that storage at elevated temperature reduces fruit quality regardless of variety. The presence of Ethysorb™ was beneficial at both storage temperatures and resulted in more quality parameters attaining the desired standard.

In general, a reduction in packaging film permeability was accompanied by an increase in the acceptability of the corresponding fruit. Under some storage conditions the incidence of *Botrytis* was the only parameter on which the packaging film failed. The only acceptable ratings were furnished by fruit packaged under PA160 film.

Summary There is clearly considerable scope to extend the quality and shelf-life of perishable fruits such as strawberries and raspberries by developing and

Taste (Score)	Colour (Score)	Firmness (%)	Weight Loss (%)	Mould (%)	PAR
Sweet (4-5)	Red (5)	81 - 100	0 - 2	0 - 1	Good (above-PAR))
Sweet/Sour (3)	Dark Red (3-4)	61 - 80	3 - 5	2 - 3	Acceptable (PAR)
Sour/Acid (1-2)	Very Dark Red (1-2)	< 60	> 5	> 4	Not acceptable (below-PAR)

For each treatment fruit-quality attributes were assessed and assigned one of three character descriptions. Each description corresponds to a 'predicted acceptability rating' (PAR).

Table 2 Classification of packaged fruit on the basis of taste, colour, firmness, weight-loss and incidence of *Botrytis*.

Variety	Storage Temp.(°C)	Storage Ethysorb™	Film type None	Film type OPP	Film type PA160	Film type PA220
Glen Lyon	15	-	C, F, W, B	F, B	T, B	T, C, F, B
		+	C, F, W, B	B	T, B	T, C, B
	4	-	W, B	B	B	B
		+	W, B	B	*	B
Glen Magna	15	-	C, W, B	B	C	C, F, B
		+	B	C	C	C, B
	4	-	C, B	B	B	T
		+	W, B	B	*	B

Each letter denotes the quality attribute for which a treatment failed to be acceptable, and is defined as: T - taste, C - colour, F - firmness, W - weight loss and B - Botrytis. Packaged fruit which had a high quality after storage is indicated '*'.

Table 3 Packaging treatments classified according to failed quality attributes.

evolving modified atmosphere packaging strategies that fully exploit the potential of both packaging films and gaseous storage regimes at harvest and during the storage period. The ability to extend shelf life has clear commercial implications but also offers unique opportunities to manipulate and dissect the control of processes which regulate ripening and post-harvest performance. This is highly complementary to existing strategies based on GM crops.

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The use of gas chromatography-mass spectrometry in the study of plant and insect defence compounds

G.W. Robertson, T. Shepherd & D.W. Griffiths

In the natural world, we are surrounded by a myriad of chemicals, some of which we can detect by our senses of taste and smell. Plants contribute much to this olfactory environment with distinctive vegetative and floral aromas and, indeed, many plants, such as the sweet pea (*Lathyrus oderatus*), are cultivated almost solely for their fragrance. Until comparatively recently, the chemical constituents of this complex environment remained unidentified and their underlying biological significance was largely ignored. The advent of sophisticated sampling and analytical techniques is changing this situation rapidly.

The involvement of chemicals in the external interactions between individual organisms is now beginning to be recognised. While most people are familiar with the concept of intra-specific (within species) communication, typified by insect sex-attractants or aggregation pheromones, it is only recently that the subtle complexities of inter-specific (between species) communication have been recognised. Plant 'info'-chemicals can affect not only neighbouring plants of the same species, but can also act indirectly on other organisms, such as insect pests, by eliciting changes in physiological and/or behavioural responses at some distance from the plant.

It is apparent that compounds that convey information over a distance between organisms must be reasonably volatile and capable of inducing a response at very low concentrations. These characteristics, together with their structural diversity, have made gas chromatography-mass spectrometry (GC-MS) an ideal technique to characterise, identify and quantify such chemicals. The advent of capillary columns with high resolving capacities has also aided in their separation from the complex mixture of compounds frequently released by insects and plants.

Part of the Phytochemistry Unit's research remit is to explore the nature of the chemical interface of various plant and insect species with their immediate biological environment, and in the course of these investigations, several examples of these 'info'-chemicals have been detected and characterised by mass spectrometry.

Pheromones released by insects can act to promote aggregation, as an oviposition stimulus, or as an alarm signal. A commonly cited example is the aphid alarm pheromone (E)-β-farnesene, a sesquiterpene hydrocarbon. Some aphid species release secretions containing this compound from their cornicles when alarmed or irritated, causing other individuals to disperse [1]. Consequently, plants that either continually release this compound or do so in response to damage, may confuse the aphids and thus reduce levels of infestation.

Plants under attack by pathogens or insects often produce elevated levels of methyl jasmonate, which appears to activate genes responsible for the production of a range of defensive compounds. The external application of this compound to plants has been shown to induce the synthesis of protease inhibitors and lycopene in tomatoes, glucosinolates in brassicas and glycoalkaloids in potatoes. This would suggest that this volatile compound may be an important means of communicating impending attack from

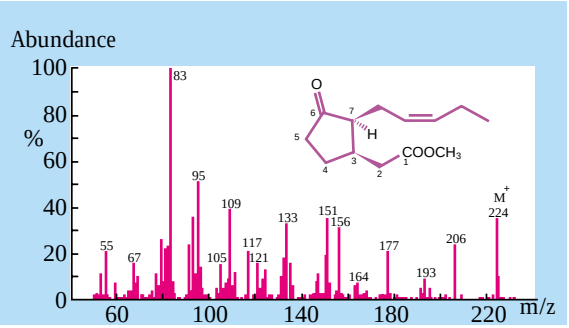

Figure 1 Mass spectrum of (+)-(3R,7S)-epi-methyl jasmonate, $C_{13}H_{20}O_3$.

plants already under threat to nearby uninfected plants, thus allowing the latter to induce their natural defence mechanisms prior to actual attack. The configuration of chiral centres in the structure of jasmonic acid and methyl jasmonate molecule at C-3 and C-7 are believed to be critical for this activity and naturally occurring jasmonates from plants are reported to have the R stereochemistry at C-3 and either the R or S at C-7. The initial isomer formed in the plant is believed to be the (+)-(3R, 7S)-form with *cis* side chains, (+)-epi-jasmonic acid, but it has a tendency to epimerize to the more thermodynamically stable (-)-(3R,7R)-form (-)-jasmonic acid with *trans* side chains [2]. In a recent study of the surface chemistry of blackcurrant (*Ribes nigrum*) leaves at SCRI [3], both epimers of methyl jasmonate were identified on the basis of their mass spectra. However, the relative concentration of the two epimers was dependant on the extraction technique employed. Solvent extraction at room temperature produced more (+)-epi-methyl jasmonate (Fig. 1), whilst steam distillation resulted in a greater proportion of (-)-methyl jasmonate, thus emphasising the importance of developing suitable sampling procedures in such investigations.

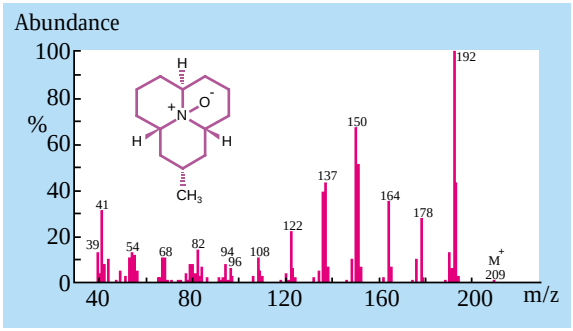

Figure 2 Mass spectrum of a mixture of precoccinelline, $C_{13}H_{23}N$ and coccinelline, $C_{13}H_{23}NO$.

Plants have other allies in their struggle against predators and among these are the coccinellid beetles, more familiarly known as ladybirds. Ladybirds are known to consume large numbers of aphids in a day and are a significant factor in reducing plant damage. They are themselves the subject of attack and have developed powerful chemical defensive weaponry of their own. The primary line of defence is a process known as reflex bleeding, where in response to the attentions of a predator, a defence fluid is exuded from the insect's knee joints. Bitter-tasting alkaloids present in the fluid make the ladybird unpalatable, and will often induce the predator to release its intended victim. The seven spotted ladybird, *Coccinella septempunctata*, produces precoccinelline and its N-oxide, coccinelline (Fig. 2), which are particularly distasteful to birds. The smaller two-spotted ladybird, *Adalia bipunctata*, produces a different alkaloid, adaline (Fig. 3), which is based on a completely different ring structure [4]. Adaline is much less unpleasant than the coccinellines, and therefore the beetle may have to exude up to seven times more alkaloid in its reflex fluid than its seven spot-

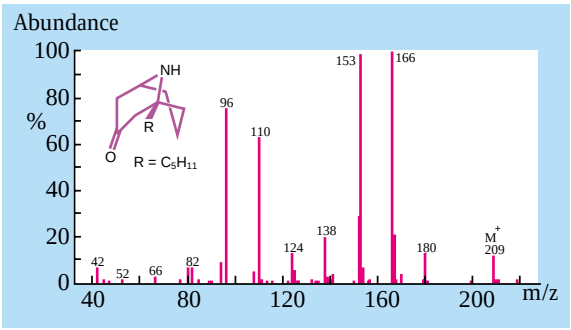

Figure 3 Mass spectrum of adaline, $C_{13}H_{23}NO$.

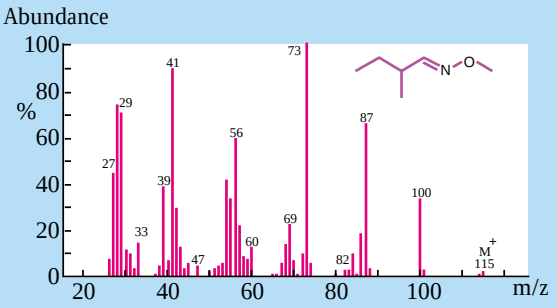

Figure 4 Mass spectrum of (E)-2 methyl butanal-O-methyl oxime, $C_6H_{13}NO$.

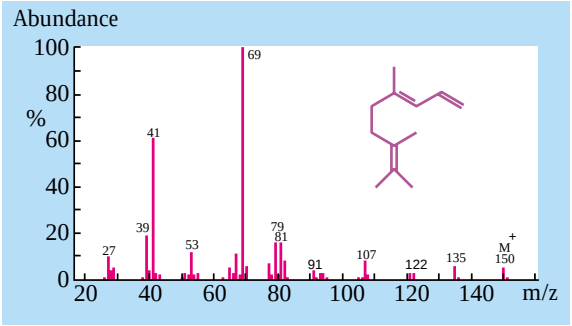

Figure 5 Mass spectrum of (E)-4,8-dimethyl-1,3,7-nonatriene, $C_{11}H_{18}$.

ted relatives, to provide an effective defence. Both types of alkaloid are aliphatic nitrogen-containing heterocycles, and are probably derived from amino acids. They were detected at SCRI in a study of lipid mobilization during parasite infestation. Production and excretion of defensive fluids is energy demanding, and the ladybirds have evolved additional defensive mechanisms. One is the bright aposematic (warning) coloration for which the beetles are renowned. Another is the emission of volatile repellent compounds, 2-methoxy-3-alkylpyrazines, based on an aromatic nitrogen-containing heterocyclic ring system.

The complexity of the (interspecific) interaction between plants and pests is well illustrated by the action of the herbivore-induced synomone given off by cucumber leaves under attack by the herbivorous spider-mite, *Tetranychus urticae* [5]. The volatile compounds given off included the homo-monoterpene (3E)-4,8-dimethyl -1,3,7-nonatriene, the acyclic monoterpene (E)-β-ocimene and (E)-2-methylbutanal, O-methyloxime. The interaction of these compounds remains unclear but (3E)-4,8-dimethyl -1,3,7-nonatriene acts as attractant to the predatory mite, *Phytoseiulus persimiliis*, which in turn reduces the herbivore mite population.

In conjunction with the Department of Soft Fruit and Perennial Crops, gas chromatography-mass spectrometry is currently being used to study raspberry (*Rubus idaeus*) flower volatiles. The larvae of the raspberry beetle, *Byturus tomentosus*, can cause significant economic damage to developing raspberry fruit, with adult beetles attracted to the flowers in early summer to congregate, feed and mate. In an endeavour to characterise the chemicals acting as attractants, flower volatiles from four commercial raspberry varieties were adsorbed onto Tenax-TA and analysed by thermal-desorption GC-MS. All four varieties were found to contain the (Z) and (E) isomers of 2-methylbutanal, O-methyloxime (Fig. 4) and 4,8-dimethyl-1,3,7-nonatriene (Fig. 5) with the (E) form predominating in each case [6]. Polyenes similar to the nonatriene have been identified as aggregation pheromones for the Australian sap beetle *Carpophilus davidsoni* [7]. The true 'info'-chemical significance of the presence of these compounds in raspberries is currently under investigation at SCRI using both an electro-antennogram linked to a GC and complementary behavioural assays.

It is clear from the examples presented that capillary gas chromatography combined with mass spectrometry is a powerful tool in revealing the complexity of the chemical environment in which both plants and insects co-exist.

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Host pathogen interactions and crop protection

Peter Palukaitis, James M. Duncan & David L. Trudgill

Plant pathology continues to play an important role in the interactions between plants and their environment, and in developing strategies for disease management. The focus of plant pathology research at SCRI continues to encompass the four broad areas of diagnosis, epidemiology, control, and plant-pathogen interactions, at the population, organismal, cellular and molecular levels. Very exciting progress has been made over the last year in a number of areas. These will be described in this section. Some of those achievements will be described in detail in selected articles, while other results and conclusions will be summarised below.

Diagnosis and epidemiology The investigation of the main sources of contamination of potatoes by *Erwinia carotovora* subsp. *atroseptica* (*Eca*) in seed and ware production, using a variety of molecular fingerprinting techniques, has been funded jointly by SOAEFD and the BPC, as a new FF project. Preliminary results show that particular isolates out of mixtures of strains soon predominated on tubers. Whether this is due to their saprotrophic or pathogenic abilities, remains to be determined. The project relies strongly on both a quantitative PCR test for *Eca*, which can detect fewer than 100 bacteria, as well as a simple and rapid extraction procedure for DNA. Such recently developed procedures allow the ready detection of plant pathogens in soil. Thus, over the last year, PCR tests have been developed for the blemish diseases of potato: silver scurf (*Helminthosporium solani*), black dot (*Colletotrichum coccodes*) and common scab (*Streptomyces scabies*). These tests can

detect c. 3 spores of each pathogen per gram of soil. Other tests include a competitive PCR for use with soil and tubers for powdery scab of potatoes (*Spongospora subterranea*), as well as several formats for PCR detection of strawberry red core (as part of the EU project REDCORE). More details on the detection of potato pathogens are contained in the specific article by D. Cullen and K. Bell.

The EU-REDCORE programme has benefited from the completion of a molecular phylogeny for *Phytophthora* based on the sequence from the internal transcribed spacer (ITS) regions of ribosomal DNA (rDNA). This valuable database, which now includes data on c. 300 isolates representing 56 species, has been used to confirm new species such as *P. quercina*, and to spot natural hybrids with potentially new host ranges, such as the *Phytophthora* spp. causing alder decline throughout Europe. This has attracted considerable interest and commercial contracts have been awarded already for the routine identification of unknown isolates using the database, as well as for characterisation of important collections of tropical species, with the results to go on the World Wide Web for use by tropical plant pathologists.

Molecular and biological analyses indicate that *Globodera pallida* (the white potato cyst nematode – PCN) is more variable in the UK, and consequently, will be much more difficult to control with resistant cultivars than *G. rostochiensis* (yellow PCN), which it has replaced. Populations of *G. pallida* most commonly contain a wide spectrum of virulence genes, so that none of the resistance genes being used by plant breeders is completely effective. Virulence genes that occur at even a low frequency will be very numerous, since the populations of PCN are immense – 10 eggs of PCN per gram of soil is equivalent to 30,000 million PCN per ha.

Parthenogenesis is frequent in soil nematodes, complicating the delimitation of species in groups, such as *Xiphinema americanum*, species of which transmit several plant viruses. An RFLP analysis of DNA from populations sampled around the world revealed at least 11 groups, confirming the distinctness of some species. However, even within recognised species clusters, substantial morphological variation was evident. It was confirmed that *X. americanum, sensu stricto* is restricted to the eastern USA, but other species were more widely distributed, e.g., populations from Crimea, South Africa, Slovakia, and the Moscow region of Russia formed a cluster identified as *X. taylori*.

In the UK, trichodorid nematodes transmitting *Tobacco rattle virus* (TRV), causing spraing in tubers, are an increasing problem, but because the incidence of spraing cannot be predicted, nematicides are widely applied as a precautionary measure. To improve detection, the ITS of the rDNA was analysed and shown to provide the basis for the rapid identification of trichodorid species. To detect TRV in the nematodes, RT-PCR has been tested and shown to be a highly sensitive method, detecting TRV even when as little as 1% of the population is viruliferous.

Chlorogenic acid, at concentrations that might realistically be found in potato tuber extracts, was found to be a potent inhibitor of PCR. Chlorogenic acid is known to be distributed unevenly in tubers and the concentration may be increased on infection. Therefore, previous reports on the erratic distribution of TRV in infected tubers, based on RT-PCR results, may, in part, be due to the uneven distribution of inhibitor(s).

A screen of 13 potato cultivars showed the presence of TRV in leaves and/or roots of plants from 11 cultivars. Moreover, virus was detected in tubers of eight cultivars, although only two (Pentland Dell and Maris Bard) developed spraing symptoms. Plants generated from the infected symptomless tubers produced symptomlessly infected daughter tubers, for the three generations tested. These daughter tubers were shown to be sources of acquisition of TRV by nematodes. Thus, movement of symptomlessly infected seed tubers may result in the dissemination of TRV and its introduction into previously unaffected sites. Recent work at SCRI has demonstrated that the virus can have a significant effect on yield and quality attributes of some of these symptomlessly infected cultivars[1].

Epidemiological studies with the virus complex responsible for groundnut rosette disease demonstrated spatial and temporal separation in the transmission of *Groundnut rosette virus* (GRV) vs. *Groundnut rosette assistor virus* (GRAV). In addition, resistance to GRV and GRAV and their aphid vector was found to vary, depending upon both age and inoculum dose, with different potentially-resistant cultivars showing different responses.

scFV antibodies have been generated against *Potato leafroll virus* (PLRV) and also to a synthetic antigen related to *Tomato yellow leafcurl virus*, for use in diagnostics and standardisation.

Other articles in this chapter focus on the variation among aphid vectors of PLRV, (Woodford *et al.*), and

on a comparison of different expression systems for production of antibody fragments for use in diagnosis or control (Zeigler *et al.*).

Control Knowledge of the host selection on fungal pathogen variation has been applied in the development of cultivar mixtures of malting quality barley, leading to smaller inputs of fungicides for control of fungal diseases. The science base for this approach and its practical application are described in the article by Newton and Swanston.

A computer-based program to aid management of *G. pallida* is well advanced. Its use has already demonstrated the difficulties of controlling *G. pallida* vs. *G. rostochiensis*, and the crucial value to farmers of having a range of commercial, resistant cultivars effective against *G. pallida* (Fig. 1). To prevent populations of *G. pallida* from increasing, the computer programme emphasises that granular nematicides are most effective when used to treat populations while they are small (< 1 egg/g soil)(Table 1). Large populations (> 50 eggs/g soil) are difficult to decrease without resorting to a fumigant followed by granular nematicides, or by rotations, which may have to be extremely long because of the slow rate of decline of some populations (Fig. 1).

The root-knot nematodes (RKN, *Meloidogyne* spp.) are the most serious nematode pests world-wide, because of their wide host ranges and ability to have several generations on one crop. This makes them difficult to control by crop rotation. As an alternative to chemical control, a collaborative EU project, co-ordinated by SCRI, examined the potential of the bacterial biocontrol agent *Pasteuria penetrans*. Laboratory assays showed that isolates of the bacterium varied in their ability to bind to and infect populations and species of RKN. Field trials in Tanzania and Ecuador showed that introducing an exotic isolate resulted in a rapid increase in spore populations and suppression of RKN infection and damage. However, this effect was observed only at sites where *P. penetrans* was already present, and the bacterium did not increase in the control plots to which the exotic isolate was not added.

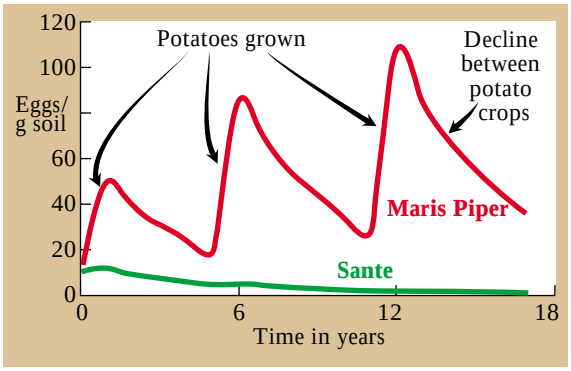

Figure 1 Changes in numbers of eggs of *Globodera pallida* per g soil in a 6 year rotation growing either susceptible cv. Maris Piper or partially resistant (80% resistant) cv. Sante, both treated with a granular nematicide which was 80% effective. Between potato crops the *G. pallida* population declines by 20% per annum.

At planting 1999	1999 Post harvest		2005 At planting	
	With nematicide	Without nematicide	With nematicide	Without nematicide
1	10.5	51.1	3.4	16.7
10	99	377	32.3	123.5
100	558	564	183	185

Table 1 Effect at planting susceptible cv. Maris Piper of differences in numbers of *Globodera pallida* (eggs/g soil) on numbers at harvest and 6 years later, for crops treated and untreated with a granular nematicide which was 80% effective. Between potato crops the *G. pallida* was assumed to decline by 20% per annum.

The TRV vector was used to express the snowdrop lectin (GNA2) in the roots of test plants. The expressed GNA2 lectin caused a reduction in the number of galls formed when plants were challenged with the RKN.

Transgenic plants expressing the coat protein (CP) gene of *Potato mop-top virus* (PMTV) showed resistance to PMTV in cvs Saturna and Pentland Marble, whether inoculated mechanically or via soil containing the infective powdery scab fungus vector of PMTV. The transgene prevented the infection of the tubers and development of spraing symptoms (PMTV and TRV both cause spraing).

In collaboration with colleagues at the National Institute of Biology, Ljubljana, Slovenia, the potato cv. Igor, particularly sensitive to the potato tuber ring necrosis disease caused by the NTN strain of *Potato virus Y* (PVY), was transformed with the CP gene of this strain. Some transgenic lines were identified which were immune to infection following graft inoculation.

A separate article in this chapter describes the integrated pest management of insect pests (Gordon *et al.*).

Plant-pathogen interactions The work on isolating potato genes involved in resistance to potato late

blight (caused by *Phytophthora infestans*) continues. Sequencing of c. 100 clones from a subtracted cDNA library, comprising 4,000 clones, has led to the identification of about 40 genes, 60% of which are involved in defence and stress reactions, and especially cell signalling. A library has also been constructed that is enriched for potato genes involved in rapid responses to infection by *Eca*. Again, many signalling genes and transcription associated factors have been identified from among c. 100 clones so far sequenced. Some of the same genes are also up-regulated upon infection with *P. infestans* in both compatible and incompatible interactions. The results obtained are described in detail in the accompanying article by Birch *et al*.

Many enquiries have been received regarding the application of this approach to other diseases, including cereal pathogens, and to the regulation of genes activated by the application of defence elicitors. Commercially-supported work on the development of elicitors as a practical control measure has also made good progress.

Complementing the work on gene expression in potato is a new core programme on the genomics of the major potato pathogens *P. infestans* and *Eca*. The development of a system for studying differential gene expression in *Eca*, allows studies on its gene regulation during the infection process. Thus, bacterial artificial chromosome (BAC) libraries are being developed for both potato pathogens. The work on *P. infestans* is part of an international *Phytophthora* Genome Initiative co-operation.

Wild tomato species have been shown to be potential sources of resistance to *G. pallida*. One such gene is the *Hero* gene, which confers about 80% resistance to most UK populations of *G. pallida*. In collaboration with M. Ganal (Gatersleben, Germany), work on the cloning of the *Hero* gene continues. However, due to changes in UK priorities for funding, no further work is continuing on searching other *Lycopersicon* spp. for new sources of resistance effective against a greater proportion of *G. pallida* populations.

The novel, multi-partite structure of the mitochondrial genome (mtDNA) of *G. pallida* has been confirmed. Instead of all the genes required for mtDNA function being contained within a single circular molecule, they are divided between several smaller molecules, each of which contains only part of the total genome. The results of sequence analysis of one of these mtDNAs and the implications of such mtDNA populations are described in an article by Armstrong *et al.*

Host resistance to nematodes may be increased by blocking the functions of nematode 'defence' genes. A screening programme of large numbers of small proteins (peptides) from a 'peptide display library' for their ability to bind to PCN secretions has detected two peptides, which bind to PCN thioredoxin peroxidase. Nematode cuticular proteins may also be involved in various recognition processes, including host resistance. Information from the international project completing the sequence of the entire genome of the free-living nematode *Caenorhabditis elegans* has been used to isolate three members of a *G. pallida* collagen multi-gene subfamily. Their expression in transformed *C. elegans* is being studied using constructs containing a 'reporter' gene using collagen promoters from both *G. pallida* and *C. elegans*.

Electron microscopy studies of roots infected by wildtype and RNA 2 mutants of the tobravirus *Pea early browning virus* (PEBV) revealed that PEBV was able to invade all tissue types in the roots, including the root tip and lateral root meristems. Whereas wildtype PEBV formed roughly spherical clumps of aggregated virus particles, mutants lacking the 2b gene (encoding a 29K nematode transmission factor) aggregated as extended columns of virus particles (Fig. 2). This

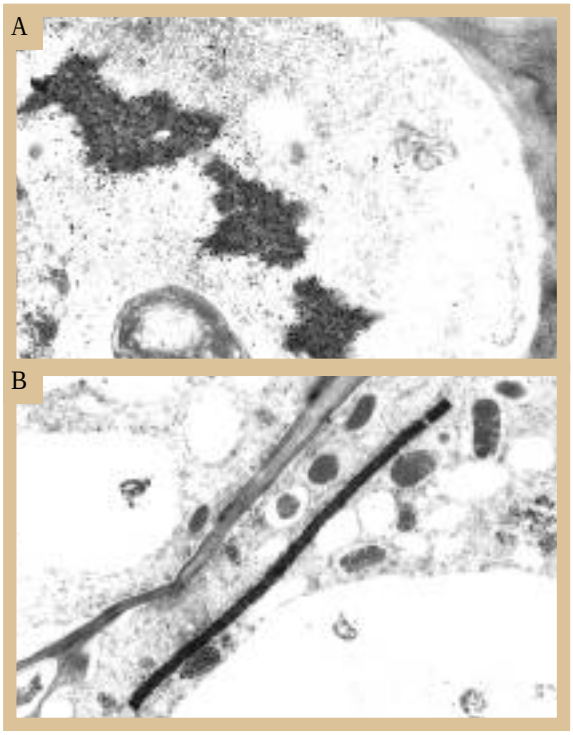

Figure 2 Electron micrographs of PEBV particles in root cells from wildtype PEBV (A) and PEBV lacking the 2b gene (B). Note the differences in aggregation profiles.

change in aggregation structure may explain why PEBV 2b mutants can no longer be transmitted by root feeding nematodes.

Expression vectors have been constructed for all three tobraviruses: TRV (described in the 97/98 Annual Report), PEBV (expressing foreign genes from a TRV CP subgenomic promoter), and *Pepper ringspot virus* (expressing from a PEBV CP subgenomic promoter). These different vectors allow the expression of non-viral proteins in a wide range of plant species.

The TRV isolate that breaks resistance in potato cv. Bintje also breaks resistance in cv. Arran Pilot, but not in cvs Record, Saturna, or Climax. (Record is one of the parents of both Saturna and Climax.) This suggests that there may be two distinct genes for resistance to TRV in cultivated potatoes.

Studies on the mechanism of CP-mediated, transgenic resistance to PMTV have revealed that the replication of RNA 3 (encoding the CP gene) was inhibited, while replication of RNAs 1 and 2 was not. Surprisingly, RNAs 1 and 2 of PMTV were able to systemically infect both the 'resistant' plants, which are resistant to the disease induced by PMTV, as well as non-transgenic plants, after sap inoculation from the resistant plants. This demonstrates both that CP is not needed for systemic movement and accumulation of PMTV RNAs 1 and 2, and that the resistance mechanism is targeted against the RNA (3) encoding the CP gene. Moreover, the accumulation of CP and its mRNA in the transgenic plants does not support a mechanism of resistance *via* virus-induced gene silencing, thus suggesting that another, potentially novel, mechanism of resistance is operating in these transgenic plants.

The mechanism of resistance to *Cucumber mosaic virus* (CMV) in transgenic tobacco expressing CMV RNA 1 was also analysed. A number of factors all argue against a virus-induced gene silencing model as the mechanism of resistance. These include: the level of expression of the transgene; the presence of a functional transgene translation product; the absence of induced resistance in a grafting experiment involving transgenic (susceptible) scion and transgenic (resistant) rootstock; the inability of the CMV-resistant, transgenic plants to inhibit the replication of *Potato virus X* (PVX) expressing CMV RNA 1 sequences; and the ability to break the resistance in transgenic scion grafted to susceptible, infected rootstock. This again suggests the operation of a novel form of resistance, but one similar to that observed in transgenic plants expressing a defective form of CMV RNA 2.

Tobacco and potato plants transformed with a full-length copy of the PLRV genome were found to accumulate virus particles in phloem cells, as do non-transgenic plants infected with PLRV by its aphid vector. Although PLRV accumulated to high levels in transgenic potato plants, comparatively little virus was found in the transgenic tobacco plants. Some of the characteristics of the transgenic tobacco plants resemble those of plants with post-transcriptional gene silencing (PTGS). The specificity of this fascinating system is described in the article by Barker *et al.*

GRV continues to produce surprising results. The ORF4 protein of GRV was able to localise to plasmodesmata when expressed from the PVX vector, while the ORF3 protein localised to the nucleolus. GRV ORF4 was able to promote CP-independent cell-to-cell movement of CMV, when expressed in place of the movement protein gene, while long-distance movement of the hybrid virus required CMV CP in the common host *Nicotiana benthamiana*, but did not occur in the CMV host *N. tabacum*. GRV ORF3 did not promote long-distance movement of CMV when it replaced the CMV CP gene, but did promote the long-distance movement of TMV, in place of the TMV CP gene, although only in the common host *N. benthamiana*. Thus, GRV ORF3 encodes a host-specific long-distance movement factor.

Thus, while we adhere to the tenets of Tennyson ("let knowledge grow from more to more", and "to follow knowledge like a sinking star, beyond the utmost bound of human thought"), the more we learn, the more surprised we become about the numerous levels and strategies of interactions between plants and their pathogens. Hence, we have to agree with Socrates, who said the more we know the more we realise how little we know.

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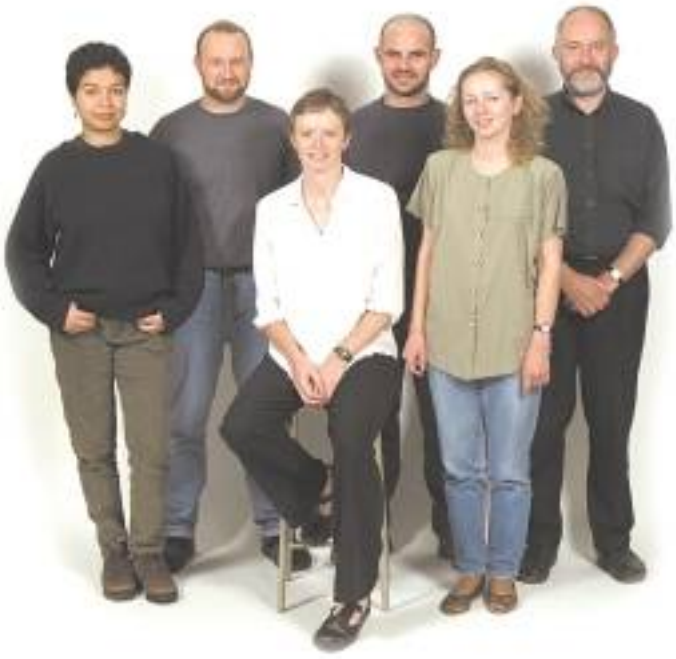
Characterising pathogen-induced signal transduction pathways in plants - opening Pandora's box

P.R.J. Birch, A.O. Avrova, A. Dellagi, J. Heilbronn, I.K. Toth & G.D. Lyon.

One of the most significant developments in biology in recent years is the world-wide commitment to fully sequence a number of genomes, including mammals such as human and rat, plants such as *Arabidopsis thaliana* and rice, and microbial genomes such as *Escherichia coli* and yeast. To date, some 21 genomes or chromosomes have been published and a further 83 microbial genomes are being sequenced. Many biotechnology companies are keen to exploit these new resources by isolating genes or expressed sequence tags (ESTs), identifying a function, initially through comparison with known genes in international databases, and exploiting novel applications with their use. One important focus of such genome studies in both plants and animals is to gain an understanding of resistance to invading pathogens. With the recent development of new molecular techniques to target the isolation of ESTs specific to a particular tissue, developmental stage or process, it is now possible to better characterise signal transduction pathways involved in disease resistance.

We are currently working to characterise the molecular bases of resistance to two important potato pathogens, *Phytophthora infestans*, the cause of late blight, and *Erwinia carotovora* subsp. *atroseptica* (*Eca*), the cause of blackleg. *P. infestans* is a fungal pathogen that has an initial biotrophic phase, later becoming necrotrophic. It shows a high degree of specificity in that it occurs as distinct races which elicit a hypersensitive response (HR) in potato cultivars possessing appropriate major resistance (R) genes. In contrast, *Eca* is a bacterial pathogen that does not show the same high degree of specificity with potato, does not occur as distinct races, and does not induce an HR. A study of plant signalling in response to infection by these two quite different pathogens enables us to obtain a better understanding of many aspects of plant-microbe signalling, including the nature of specificity and factors involved in hypersensitivity. In addition, these two pathogens cause important economic diseases of potato within the UK and are amongst the three most important pathogens of potato world-wide.

Specificity in cell signalling can arise at several levels, from the receptor, through modulation of signalling kinetics, interactions of different signalling pathways, and at the level of tissue-specific downstream regulators. Thus, tissue-specific transcriptional complexes could allow similar upstream signals to regulate different sets of genes in different tissues. For this reason, if we are to fully understand the molecular bases of disease resistance, we should look at all the molecular processes associated with induced resistance.



We compared the potato cultivars Stirling, which possesses both horizontal resistance and major (R gene) resistance to *P. infestans*, and Bintje, which is highly susceptible and does not exhibit an HR. We used recently developed PCR-based methods for isolating differen-

tially expressed genes, cDNA-AFLP and suppression subtractive hybridisation (SSH), to generate a cDNA library enriched for *P. infestans*-induced genes specific to Stirling at a time-point of 24 hours post-inoculation. cDNA prepared from the infected Bintje was used as a driver to remove all common sequences from the infected Stirling via the subtraction procedures associated with SSH. The remaining material was amplified and cloned. One thousand clones prepared from this cDNA were screened by hybridisation to remove repetitive sequences such as ribosomal RNA. One hundred of the remaining clones were sequenced and compared to databases using both FASTA (DNA-DNA) and BLASTX (DNA-protein) searches. Fifty per cent matched sequences in international databases. Of these, approximately 60% showed similarity to previously characterised stress-, defense-, or senescence-associated genes of plant origin. In addition, a number of identified genes are implicated in programmed cell death, including signal transduction (serine palmitoyltransferase and phosphatidylinositol-4-phosphate-5-kinase), protein degradation (ubiquitin, ubiquitin carrier protein and cysteine protease), DNA degradation (cyclophilin) and metal ion chelation (metallothionein), and may thus be involved in the HR[1].

A similar approach, using SSH, was carried out with potato Stirling one hour after infiltration with *Eca*, using uninfected Stirling as a driver. The intention here was to generate a cDNA library highly enriched for early response genes, including those involved in signalling. Analysis of the *Eca*-induced cDNA library showed that it is highly redundant, i.e. it contains only a few different genes, and confirmed that it does indeed contain a number of genes related to transcriptional regulation and signal transduction. These include genes encoding a WRKY-like transcription factor, a protein phosphatase (PP2A) regulatory subunit, and a ubiquitin-specific protease. The expression profiles (Northerns) of these genes were studied at different times after inoculation with either *Eca* or *P. infestans* (compatible and incompatible interactions) and confirmed their early, pathogen-induced up-regulation. Other genes in the *Eca*-induced cDNA library included matches to previously reported genes of unknown function. These include a dehydration-induced gene ERD15, a phosphate-induced gene *phi-1* and a *Meloidogyne*-induced giant cell gene. Sequences matching a gypsy-like retrotransposon were also identified. No function could be ascribed to approximately 50% of the sequences as they show no similarity to known genes.

Both cDNA libraries described above contain a number of chloroplast-associated genes. This may reflect a non-specific induction, possibly through changes in redox potential, which is known to be a regulator of chloroplast gene transcription, or a more specific induction possibly involving free radical production associated with the oxidative burst.

As each isolated gene is putatively identified via database searches, it is positioned within a model of the infected plant cell (Fig. 1) which is being developed as knowledge of signalling processes emerges in the literature. This allows us to hypothesise about the roles of gene products within the plant-pathogen interaction and stimulates the design of future experiments to test the functions of these genes.

As we continue to sequence clones from these sub-

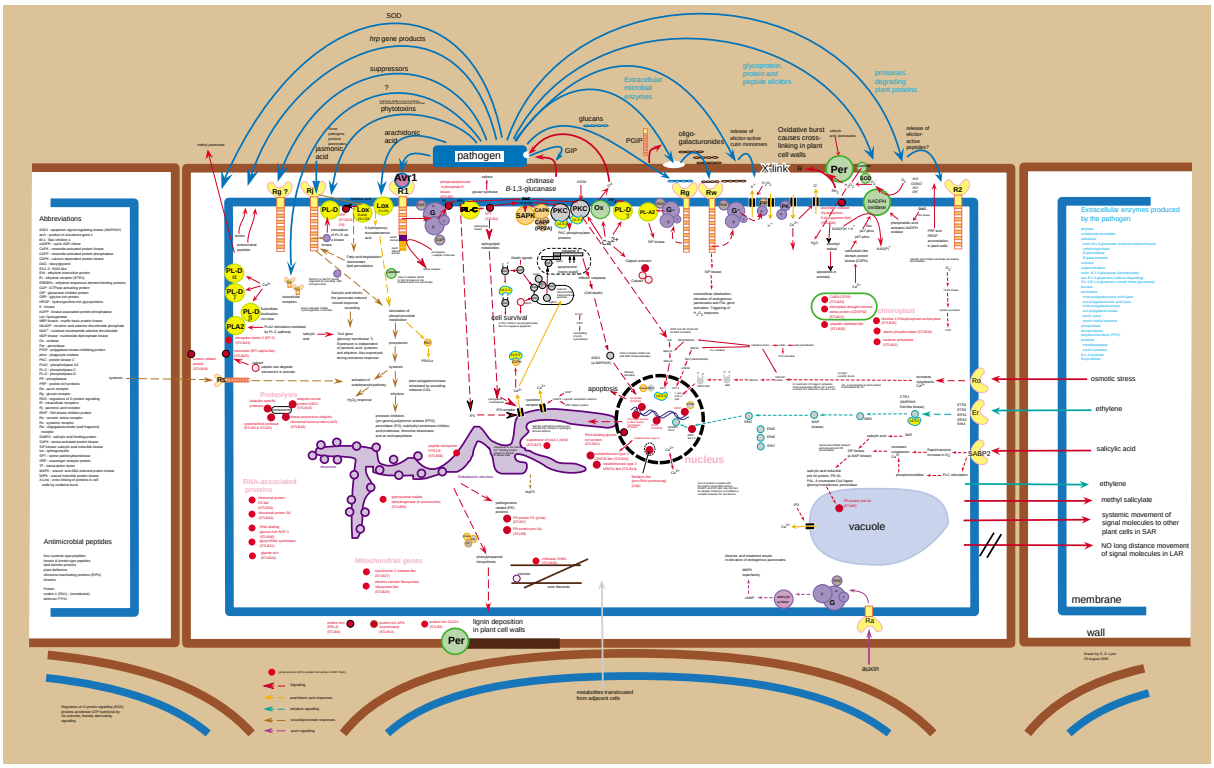


Figure 1 Cell signalling in resistance - a hypothesis.

tracted cDNA libraries, gain more information about their profiles of expression, and identify their function through, for example, transformation studies, we hope to form a picture of the signalling pathways involved in resistance to late-blight and in the HR in general. With an understanding of such processes, there will be increased potential to engineer resistance cascades in order to modify resistance responses. In addition, promoters will emerge that respond rapidly to general pathogen attack and may thus be useful in strategies to engineer broad-spectrum resistance. Furthermore, as efforts to understand cell signalling increase worldwide, a picture is emerging of the considerable conservation in programmed cell death processes between plants and animals.

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Diagnostics for skin blemish pathogens of potato

D.W. Cullen & K.S. Bell

The British potato industry suffers, on average, 1 million tonnes of waste annually through disease, damage, and failure to meet market specifications. The British Potato Council (BPC) estimated the annual cost of avoidable waste between leaving the field and reaching the consumer at £30 million[1]. Providing practical solutions to wastage problems is therefore regarded as a high priority throughout the whole industry. Tuber skin blemish diseases are a major cause of wastage and these are of increasing concern due to the demand for washed potatoes with a high quality appearance for the fresh 'pre-pack' market.

The bacterial disease common scab (*Streptomyces scabies*) and the three 'fungal' diseases, powdery scab (*Spongospora subterranea* f. sp. *subterannea*), silver scurf (*Helminthosporium solani*), and black dot (*Colletotrichum coccodes*), are the predominant blemish diseases in the UK. These diseases primarily reduce the market value of potato crops but can also affect the yield. Annual losses from the ware crop were estimated at £3 million for common scab, £1 million for powdery scab, and up to £5 million for silver scurf and black dot, and there are additional losses in the seed industry[1]. The use of disease-susceptible varieties, spread of inoculum on seed potatoes, and changing agricultural practices have all increased the incidence of blemish diseases.

Classical symptoms for common and powdery scab include the appearance of raised or pitted corky scabs on the tuber surface. Tubers infected with silver scurf and black dot show grey or silver patches, whilst black microsclerotia may form on the tuber surface to produce a 'sooty' appearance with the latter disease (Fig. 1). Infection by these pathogens can arise from both contaminated seed tubers and soil-borne inoculum. An improved understanding of the epidemiology of these diseases will assist in their control, thus improving the quality and efficiency of ware production and ensuring supplies of healthy seed. The BPC has funded the development of molecular diagnostics for powdery scab, and MAFF has funded a similar project to study the epidemiology of the soil-borne phase of common scab, black dot, and silver scurf. The objectives were to develop rapid assays to allow specific detection and quantification of these pathogens in potatoes and soil. The diagnostic method selected was the polymerase chain reaction (PCR), which allows the exponential amplification of specific DNA fragments from complex DNA samples by *in vitro* DNA synthesis[2]. This highly sensitive procedure requires a DNA template containing the region to be amplified and two oligonucleotide 'primers' flanking this target region. The amplification process is automated by the use of a thermocycler and a thermostable DNA (*Taq*) polymerase. Another key requirement for both projects was the development of a method to extract and purify microbial DNA from soil.

Figure 1 Classical symptoms of common scab, silver scurf, black dot, and powdery scab.

This substrate presents problems because microbes can be protected from lysis in the soil matrix and co-extracted humic compounds are potent inhibitors of PCR.

Common Scab, Silver Scurf, and Black Dot The key step in the development of a successful PCR assay is the identification of DNA sequences that are characteristic for those species under study. These are used to design specific primers. For fungal pathogens, regions in the nuclear rDNA gene unit are often targeted[3]. Genus and species-specific primers have been designed in the non-coding, variable internal transcribed spacer (ITS) regions 1 and 2 of rDNA. ITS1 and ITS2 sequences of *C. coccodes* reference isolates are present in the databases, whereas these regions had to be sequenced for several UK isolates of *H. solani*. Primers were then designed to unique sequences within these ITS regions for specific detection of *H. solani* and *C. coccodes*. In the case of *S. scabies*, primers were designed to detect the proposed pathogenicity gene (*nec1*), recently described[4]. *Nec1* confers the necrogenic potential when acquired by non-pathogenic strains of *Streptomyces*.

Two sets of primers (outer and nested) were designed for *H. solani*, *C. coccodes*, and *S. scabies*. Nested PCR was used to increase specificity and sensitivity of single-round PCR, and specific product was detected for each organism when 10 fg (10^{-15} g) DNA was included per reaction, the equivalent of one genome. Positive results were obtained when 25 different *H. solani* and *C. coccodes* isolates were tested. Preliminary results also indicated that the *nec1* primers amplified product from other pathogenic but not from non-pathogenic isolates of *Streptomyces* spp. Comparisons between each primer to DNA and protein databases of other fungi and bacteria revealed no significant levels of similarity. The specificity of primers was confirmed as no PCR products were amplified when testing DNA from a range of different fungal and bacterial plant pathogens in the SCRI collection.

A simple and rapid procedure for direct extraction of DNA from soils[5] was modified and recovered high molecular weight DNA of a suitable purity for PCR within 3 hours. The key steps of the protocol were the physical disruption of soil microbes in a Mini-Beadbeater, the reduction of humic contamination by the use of an alkaline sodium phosphate + CTAB buffer, and purification of DNA extracts *via* polyvinylpolypyrrolidone (PVPP) or Sephadex G-75 spin-column chromatography (Fig. 2).

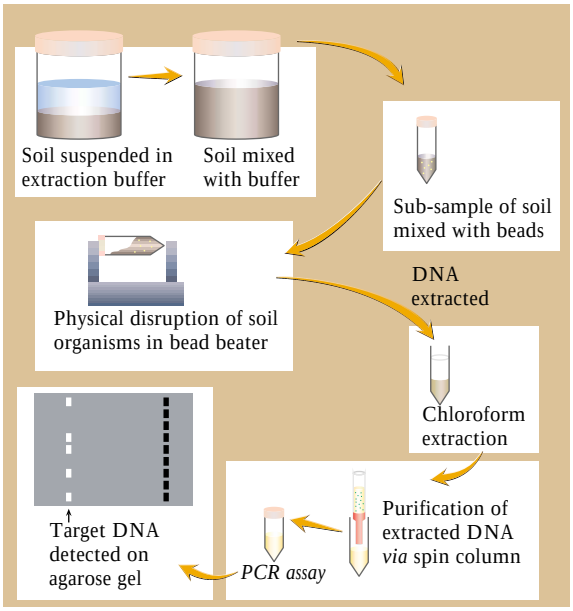

Figure 2 Extraction of DNA from soil.

Primers were designed to operate under the same conditions to ensure adaptability for a rapid multiplex PCR assay (*i.e.* incorporation of the three primer sets in one reaction), in which all three pathogens could be detected simultaneously. The sensitivity of PCR for the specific detection of *H. solani*, *C. coccodes*, and *S. scabies* in seeded soils was tested in parallel for both the single primer sets and in multiplex PCR. Soil samples were seeded with each organism, singly and in combination, and the level of sensitivity for both PCR systems was set at 3 spores per gram of soil, the lowest level of inoculum added (Fig. 3). However, multiplex PCR had the advantage of reducing the time and cost of the procedure. Work is underway to set limits of PCR detection for each organism in plant material. An automated and quantitative PCR system to eliminate the need for gel electrophoresis also will be developed.

Powdery Scab As with *H. solani*, the rDNA unit of *S. subterranea* f. sp. *subterranea* was partially sequenced and the ITS regions were compared to related organisms in the databases in order to design species-specific primers. Selected primers were shown to be highly specific as they amplified the predicted size of product (391 bp) from *S. subterranea* f. sp. *subterranea* but not from related organisms or other potato pathogens. *S. subterranea* was detected in peel and washings of diseased and apparently healthy tubers, but not in Scottish classified seed potatoes or axenically micropropagated tubers. It was possible to detect *S. subterranea* in soil at levels of 1-10 spore balls

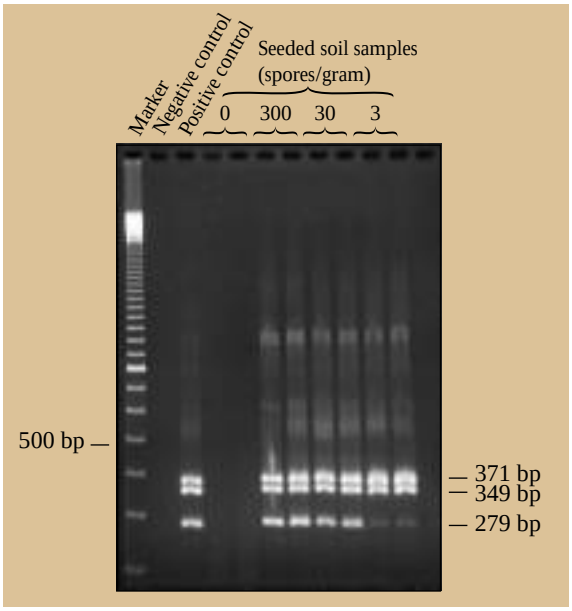

Figure 3 Detection of *H. solani* (371 bp), *C. coccodes* (349 bp), and *S. scabies* (279 bp) in spiked soil by nested multiplex PCR.

per gram with the PCR assay and aforementioned soil DNA extraction protocol. A quantitative PCR assay was also developed using an internal control (competitor) based on a DNA fragment of a smaller size (249 bp) to the target but with the same primer binding sites. A fixed concentration of 'competitor DNA' was co-amplified with DNA extracted from a dilution series of *S. subterranea* spore balls, and the ratio of the amount of both products was estimated from the intensity of the bands on a gel (Fig. 4). This data was used to generate a standard curve to estimate the DNA concentration from unknown numbers of sporeballs amplified under the same conditions, and hence the numbers of target organisms could be determined in a test sample.

PCR-based tests are useful tools for rapid and accurate assessment of tuber and soil contamination by plant pathogens, and will assist epidemiological studies. Quantitative results can be obtained in a day, in contrast to several weeks for glasshouse-based bait tests. Results will supplement our current knowledge on methods to control these pathogens (*e.g.* early harvesting, maximum levels of seed health, fungicide seed treatments, and strict hygiene in seed and ware stores) and will provide a basis for the development of new integrated management strategies. Whilst control of soil-borne diseases is inherently difficult, there are possible strategies to minimise economic loss. Quantitative data on inoculum levels would be an important component of disease risk assesments to assist in decision making on control issues such as the application of biocides; avoiding high risk fields; matching cultivars to fields according to their respective resistance rating and disease risk; crop rotation strategy; irrigation regime; and prioritising land according to the crop end use.

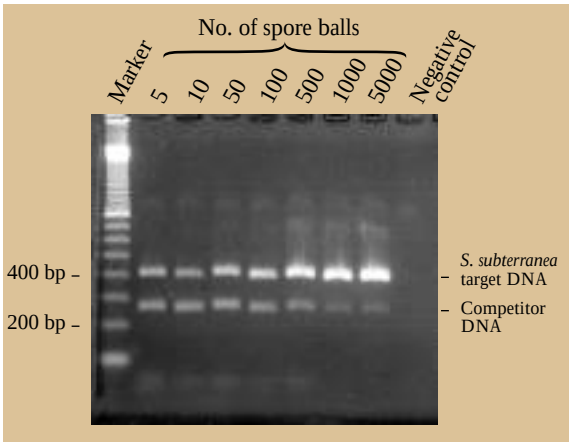

Figure 4 PCR products obtained after using primers Sps1 and Sps2 to coamplify 1fg of competitor DNA and *S. subterranea* DNA extracted from varying amounts of spore balls.

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Heterologous expression systems for the production of functional antibody fragments

A. Ziegler, B. Reavy, G. Cowan, M. Mayo & L. Torrance

Introduction Antibody fragments (scFv), either selected from phage display libraries or derived from antibody-secreting hybridoma cell lines, can be cloned and functionally expressed in heterologous systems (see Ann. Rep., 1995, 125-127). These reagents are now being used in numerous biotechnological and biomedical applications, such as diagnosis and therapy. ScFv can be a source of standardised reagents for use in plant pathology and other areas of the plant sciences. Furthermore, by expressing anti-viral scFv in their cells, plants can be made disease-resistant. ScFv that recognise molecules naturally found in plants, can be expressed in plants to immunomodulate plant metabolism.

We have produced novel diagnostic reagents for plant viruses and plant enzymes by selecting antibody fragments from phage display libraries, or by cloning genes that encode scFv from existing hybridoma cell lines. The scFv reagents have been expressed in *Escherichia coli* cells and used successfully for virus detection by various immunological methods, such as ELISA, Western blotting and tissue printing (see Ann. rep., 1997/98, 111-113). The reagents were extensively tested (EC AIR3 project CT94-1046) and found to perform as well as monoclonal or polyclonal antibodies.

Heterologous expression of scFv Antibody fragments that are to be used in disease diagnosis or therapy have to be produced in large quantities and the preparations must be of consistently good quality. This report describes scFv expression in four different systems.

E. coli

Traditionally the most widely used organism for the heterologous expression of antibody fragments, the advantages of *E. coli* include easy manipulation, rapid growth of the bacteria, and simple media requirements. An scFv (scSCR20), derived by molecular cloning from a hybridoma cell line that secretes an anti- African cassava mosaic virus monoclonal antibody, was expressed in *E. coli*. The yield in simple shake flasks was 1 mg scFv per litre of culture.

However, there are also potential disadvantages with the bacterial system. For example, loss of the plasmid during culture, or toxicity of some scFv sequences, can lead to low or no accumulation of scFv, or to the production of insoluble protein aggregates. Also, the post-translational modifications of the recombinant protein that could occur in eukaryotic cells are not possible in prokaryotic systems such as *E. coli*. Therefore, in parallel with the expression in *E. coli*, alternative methods of scFv production such as use of insect cells and plants were compared.

Insect cells

Two different systems were compared: the Baculovirus expression system (Invitrogen), which adapts an insect virus for the transient expression of recombinant protein in infected cells, and the *Drosophila* expression system (DES, Invitrogen), in which insect cell lines are stably transformed.

BACULOVIRUS SYSTEM: The *Baculovirus* vectors for the infection of *Spodoptera frugiperda* (or other insect) cells contained the genetic information for a melittin signal peptide derived from the honey bee. This sequence facilitates the secretion of the expressed protein to the culture medium. However, when scSCR20 was expressed from the *Baculovirus* vector in any of three different insect cell lines, the scFv was retained within the cells (Fig. 1) and could not be detected in the culture medium. This could be a result of the baculovirus infection affecting protein

secretory mechanisms in the infected insect cell. However, other proteins can be secreted using the baculovirus system. Probably, specific features such as the amino acid sequence of the scFv can modify the behaviour of the expressed fusion protein.

DROSOPHILA SYSTEM: In contrast to the Baculovirus system, the *Drosophila* system relies on stable cell lines that express the foreign protein. Since the *Drosophila* cells will spontaneously incorporate hundreds of copies of the transfected genetic information, the expression levels should be very high, and no extensive testing of cell lines for expression is required. Stable cell lines can be established after co-transfection with a plasmid that confers resistance to the antibiotic hygromycin B. The expression of the foreign protein is under the control of the metallothionein promoter, and the system can be induced by the addition of non-toxic concentrations of metal ions such as copper. For scSCR 20, a yield of 20 mg per litre of cell culture was obtained. The bulk of the scFv was found secreted to the culture medium (Fig. 1) as expected, which allowed easy purification of the recombinant scFv.

Plants

Plants, too, can be used as alternatives to microbial fermentation for the cost-effective production of large amounts of therapeutic proteins, including antibodies. We have investigated the effects of the expression of scFv in plants on virus replication and on plant metabolism, using scFv specific for virus-encoded replicase and a scFv that binds to a plant enzyme, potato granule-bound starch synthase (GBSS).

Figure 1 Immunoblot of scSCR20 produced in insect cells. D Drosophila system B Baculovirus system.

However, not every antibody sequence is equally well suited for high-level expression in plants. There are, as yet, no general rules for the design of effective constructs that result in accumulation of functional scFv. In order to assess quickly the performance of a number of different constructs, a transient expression system based on the plant virus, Potato virus X, was employed (see Ann. Rep., 1995, 125-127).

The system allows a variety of different constructs to be evaluated (Fig. 2) in a short time. The influence of signal sequences and fusion proteins on the localisation, level of accumulation and functionality of the product can be analysed quickly.

Using the PVX system, an scFv against GBSS that had been selected from an antibody phage display library, was expressed in *N. benthamiana* as a fusion with an IgG human kappa constant region. A murine IgG leader sequence was used to target the scFv to the apoplast. ScFv's recovered from plant extracts were found to be functional in both ELISA and Western blot for the detection of GBSS (Fig. 3). Also, a number of constructs for the expression of scSCR20 were successfully tested using the PVX system.

ScFv constructs found suitable in the PVX system were used for *Agrobacterium* mediated transformation of potato. Expression levels reached 0.5% of total soluble protein.

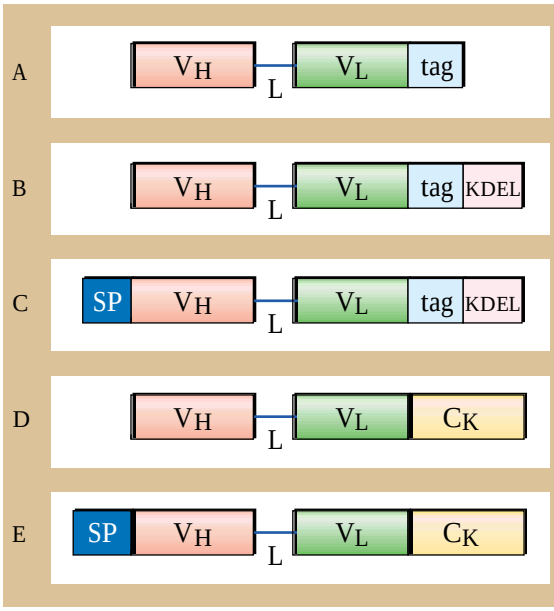

Figure 2 Structure of different scFv constructs tested in the PVX expression system.
$V_H \overline{L} V_L$ = scFv
SP = murine IgG leader
tag = peptide epitope tag for detection
KDEL = signal peptide for retention in endoplasmic reticulum
C_K = IgG human kappa constant region

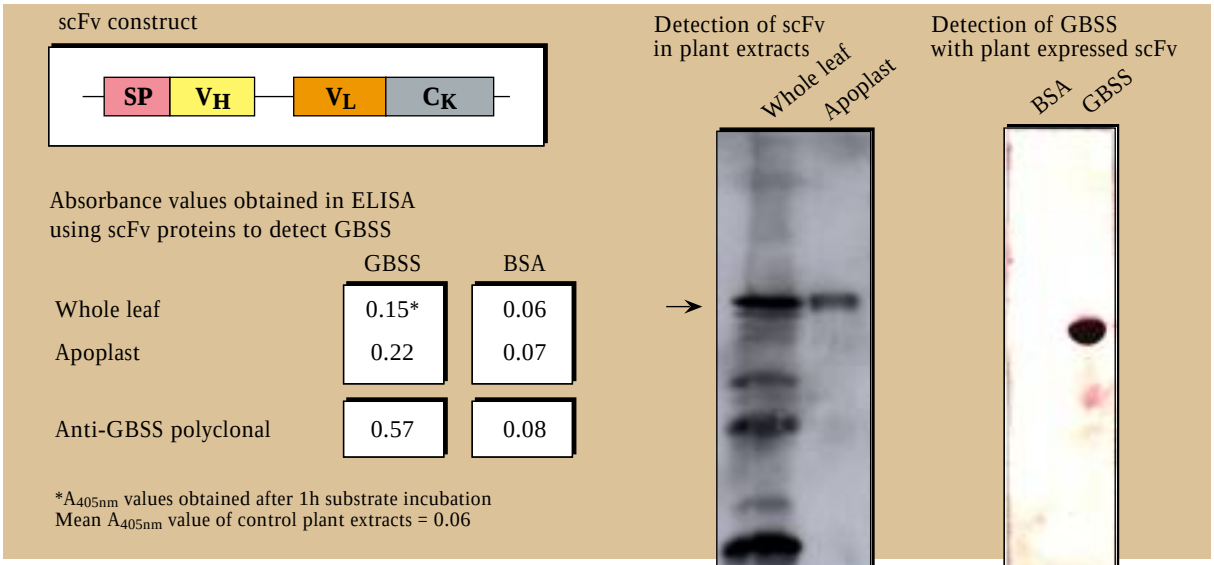

Figure 3 PVX vector expression of GBSS-specific single chain Fv.

Conclusions The choice of expression system depends on a number of factors, such as the projected use of the scFv (e.g. diagnosis, therapy or antibody mediated resistance). Also, the quantities of scFv required and the methods of purification and validation that are available for each system. Once a system has been chosen, it has to be optimised for each scFv sequence. There is no one expression protocol that is ideally suited for every scFv.

Insect cells offer a cost effective, high efficiency alternative to *E. coli* expression.

Unlike *E. coli*, insect cells are capable of performing post-translational modifications similar to mammalian cells. The insect cells can be grown in stirrer vessels at room temperature, which can help to stabilise the scFv, so there is no need for specially equipped incubators.

For scSCR20, the yield using the *Drosophila* system was 10x the yield from the *E. coli* expression. With the PVX system, testing of constructs for plant expression of scFv can be done before committing time and resources to the production and analysis of stable transformed lines using *Agrobacterium*, a process that usually takes many months.

Acknowledgements

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Plants transformed to express the entire genome of *Potato leafroll virus*

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Introduction *Potato leafroll virus* (PLRV) is transmitted in a persistent non-propagative manner by aphids that put virus into the vascular tissue of plants, where it remains largely restricted to the phloem cells. PLRV is not mechanically transmissible but plants can be agroinoculated by direct injection of *Agrobacterium tumefaciens* cells that carry a DNA copy of the virus genome in a Ti plasmid. Transcripts of the plasmid then initiate infection. As when aphids inoculate plants, the resulting infections are limited to phloem tissue. An alternative way of initiating virus replication in a plant is by stable transformation with a full-length (biologically active) cDNA copy of a virus genome, and this has been done for a number of viruses with RNA genomes. In recent work, we have transformed tobacco and potato plants with full-length infectious cDNA to the PLRV genome. These plants are providing novel and unexpected insights into the replication of PLRV and its interaction with the host genome.

Transformation of plants and characterisation of transgenic lines A full-length cDNA copy of the PLRV genome was cloned into a plasmid used for *Agrobacterium tumefaciens*-mediated transformation. The construct was used to transform tissues of *Nicotiana tabacum* cv. 'Samsun' and the potato genotypes cv. Maris Piper and the highly PLRV-resistant SCRI breeding line G8107(1). The transgene contains the PLRV sequence under the transcriptional control of the 35*S* constitutive promoter from *Cauliflower mosaic virus*, and the selectable marker gene, NPTII, that confers resistance to kanamycin.

Two transgenic lines of tobacco were obtained and seeds were collected from self-fertilised flowers. The transgene was detected in DNA extracted from T_1 seedlings by PCR. The observed segregation data of kanamycin-resistant:sensitive seedlings from lines AW3 and AW14 indicate that T-DNA was inserted at two loci in the genome of the T_0 parental plants.

It was not possible to obtain transgenic lines of Maris Piper expressing PLRV, because transformed callus grew poorly, then turned brown and died without regenerating shoots. ELISA showed that PLRV coat protein (CP) was made in the callus cells; it is possible that PLRV replication in the callus caused stress that inhibited growth and shoot regeneration.

Transformation of *S. tuberosum* clone G8107(1), which has strong host-mediated resistance to PLRV accumulation, produced vigorously-growing callus from which shoots regenerated. Plantlets of four transgenic lines (BF#lines) were transferred to the glasshouse and grown to maturity to produce a few tubers.

PLRV expression in transgenic plants

Tobacco: Many T_1 seedlings of the two transgenic tobacco lines were found to contain PLRV antigen (detected by ELISA of leaf tissue) from an early stage in their growth. Transgenic plants were visually indistinguishable from infected wild-type (wt) plants and bore no symptoms. No PLRV could be detected in approximately 25% of T_1 transgenic plants by serological tests, and PLRV RNA was not detected by hybridization tests in such plants. The concentration of PLRV detected by ELISA varied greatly among plants of the same line. Thus, plants of line AW3 with the highest concentration of antigen, contained 12-fold more than the plant with the lowest concentration, and in line

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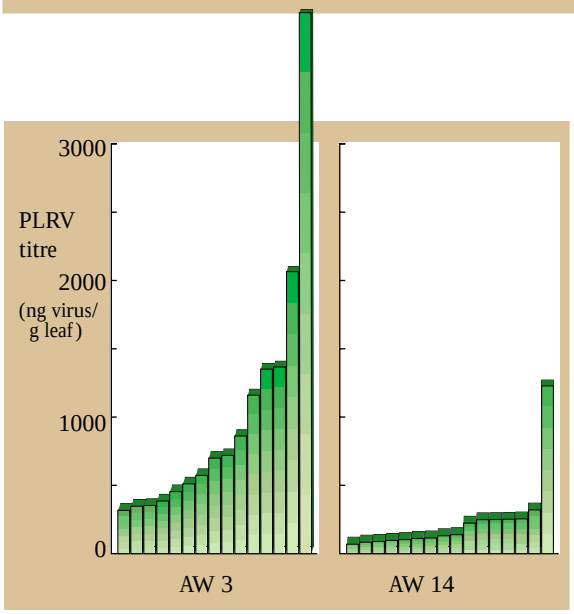

Figure 1 Detection of PLRV antigen in transgenic plants of lines AW3 and AW14, 10 weeks after sowing. Data from five AW3 and four AW14 plants that were transgenic but in which no virus could be detected, are not shown. The bars represent virus titre (ng virus/g leaf) in young fully-expanded leaves from individual plants.

AW14 the difference was 18-fold (Fig. 1). However, occasionally virus could not be detected in leaf samples of plants that produced other infected leaves, presumably because the sensitivity of the ELISA was insufficient to detect the low concentration present. The amounts of PLRV antigen in young fully expanded leaves of AW3 transgenic plants and of infected wt plants were similar; in 8 week-old plants, the mean titres were about 600 ng virus/g leaf.

Potato: Glasshouse-grown plants of BF lines initially did not look substantially different from infected wt plants of G8107(1); they did not show the 'leafrolling' symptoms characteristic of PLRV infection. However, as the plants aged, most leaves of BF plants, except those at the top of the stem, became necrotic and died prematurely and plants were severely stunted in comparison to PLRV-infected wt

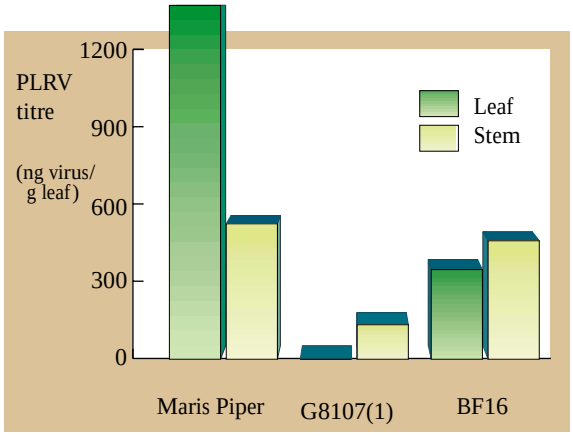

Figure 2 Detection of PLRV in leaf and stem tissue of wt infected plants of Maris Piper, line G8107(1) and transgenic line BF16.

plants of G8107(1), which continued to grow without symptoms. BF plants remained severely stunted throughout their lives and only produced a few small tubers. Plants grown from these tubers, and tested by ELISA, contained high concentrations of PLRV, whereas plants grown from tubers of infected non-transgenic G8107(1) contained very little virus (Fig. 2). More PLRV could be detected in stem tissue than in leaves of G8107(1) and BF plants, but the reverse was true for Maris Piper plants (Fig. 2). Both leaf and stem tissue of infected wt plants of the susceptible cv. Maris Piper contained high concentrations of PLRV (Fig. 2).

Properties of PLRV in transgenic tobacco plants The virus-like particles that accumulated in the transgenic plants were indistinguishable from those of PLRV from conventionally infected plants, in immunosorbent electron microscopy (ISEM) tests. The PLRV that accumulated in the transgenic plants infected all receptor plants, when scions from transgenic plants were grafted to virus-free, non-transgenic tobacco plants, and was transmissible by *Myzus persicae* to virus-free receptor plants of *Physalis floridana*, *N. clevelandii* or *N. tabacum*.

Location of PLRV by tissue printing The number and distribution of cells

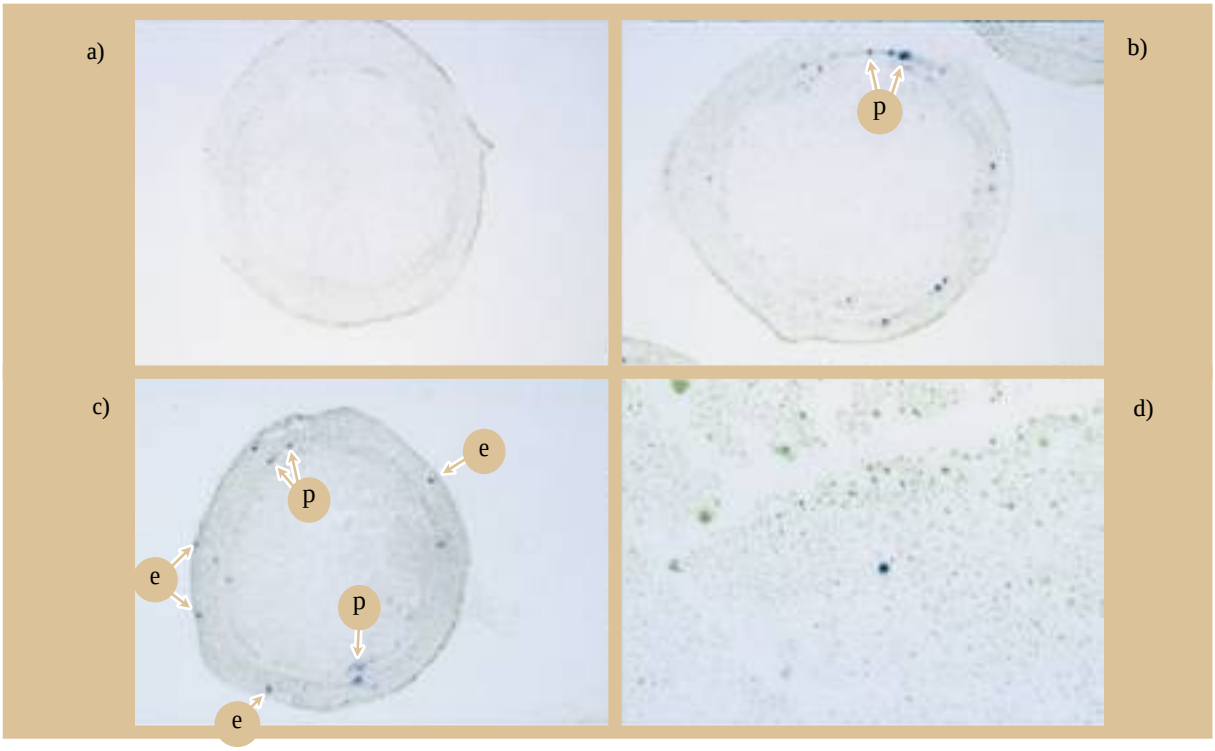

Figure 3 Detection of PLRV-infected cells in tissue prints. (a) Stem print of a virus-free non-transgenic tobacco. (b) Stem print of a PLRV-infected non-transgenic tobacco. Stained phloem cells are labelled 'p'. (c) Stem print of transgenic AW3 tobacco. Stained phloem cells are labelled 'p' and stained epidermal cells are labelled 'e'. (d) Leaf print of transgenic AW3 tobacco showing one stained cell.

containing PLRV was assessed in leaves and stems of transgenic plants by making tissue prints (immunologically stained imprints of tissues on a nitrocellulose membrane). Stained spots, indicating the location of infected cells, were readily identified in transgenic and infected wt plants, but were not seen in any of the prints made from virus-free plants.

Tobacco: Tissue prints of tobacco stem sections showed that the majority of phloem cells in transgenic and infected wt plants were unstained, with less than 5% of the phloem companion cells estimated to be infected (Fig. 3b and 3c). However, a few infected cells were also observed in epidermal tissue of stem sections from some transgenic tobacco plants (a mean of about one cell per section), but PLRV was never detected in epidermal cells from infected wt plants (Fig. 3c vs. 3b).

Tissue prints were also made of leaf lamina of transgenic tobacco from which the lower epidermis had been removed by peeling. Tissue pieces were pressed onto nitrocellulose membranes to leave an imprint of the exposed tissue. Infected 'mesophyll' cells were observed in leaves of transgenic tobacco plants (Fig. 3d) but not in infected wt plants. In some transgenic tobacco leaves, mesophyll cells were aggregated to form small clusters. Infected mesophyll cells were found in only a few tissue prints; a total of 36 infected cells were found in 37 cm^2 of leaf prints made from transgenic leaves. We estimate that about one in 40000 mesophyll cells of these leaves had accumulated detectable amounts of virus.

Potato: Tissue prints of stem sections of Maris Piper showed that many phloem cells in infected wt plants were stained, although far fewer cells were infected in PLRV-infected G8107(1) (Fig. 4b) (less than a third in comparison to Maris Piper). Many phloem cells were infected in stems of transgenic plants of BF lines.

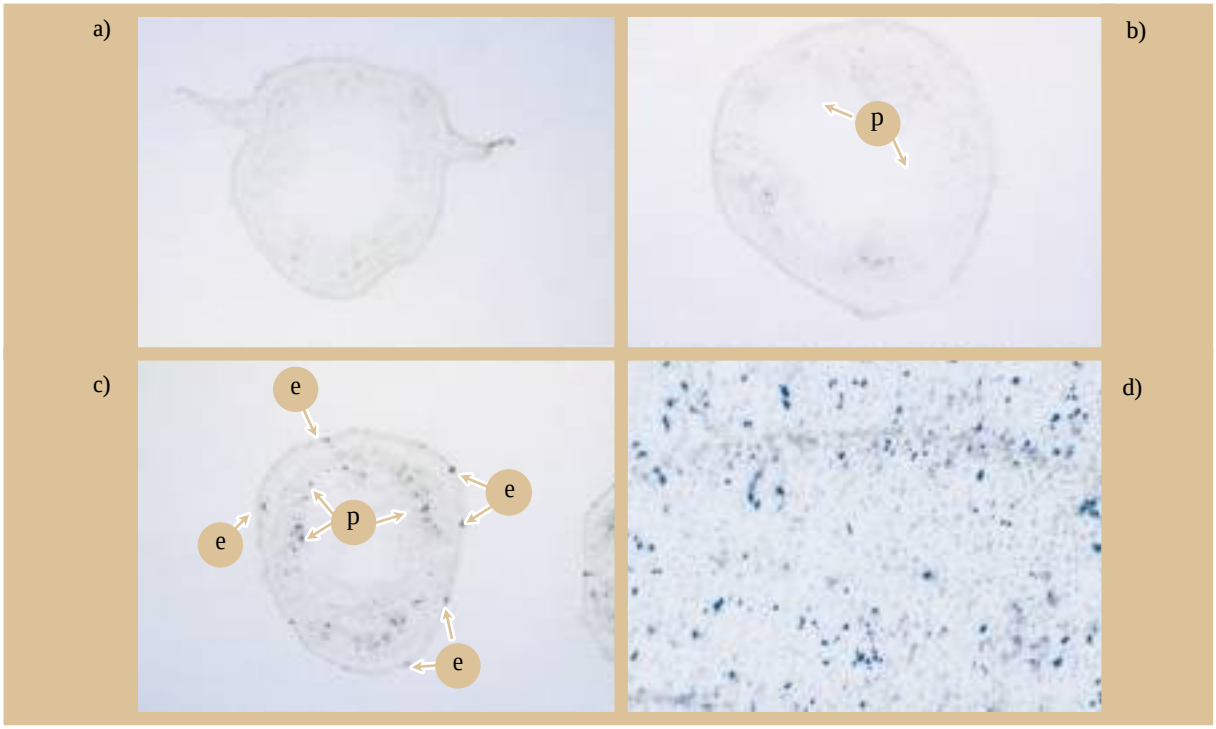

Figure 4 Detection of PLRV-infected cells in tissue prints. (a) Stem print of a virus-free non-transgenic potato. (b) Stem print of a PLRV-infected non-transgenic G8107(1) potato. Stained phloem cells are labelled 'p'. (c) Stem print of transgenic BF16 potato. Stained phloem cells are labelled 'p' and stained epidermal cells are labelled 'e'. (d) Leaf print of transgenic BF16 potato showing many stained cells.

More interestingly, a substantial proportion of infected cells also were observed in stem epidermal tissue, although PLRV was never detected in epidermal cells of infected wt G8107(1) plants (Fig. 4c vs. 4b). Tissue prints of leaf lamina of transgenic plants of BF lines showed that many cells were infected in some samples (Fig. 4d). However, the distribution of infected mesophyll cells was very erratic and some areas of leaf contained no infected cells, while neighbouring areas of the same leaf were very heavily infected.

Discussion The simplest expectation for plants transformed with cDNA copies of the PLRV genome is that all cells will produce transcript RNA and virus will thus multiply in all cells. But this did not happen with tobacco or with the highly PLRV-resistant potato clone. Thus, some mechanism was restricting PLRV multiplication in most of the cells of these plants. It is possible that the failure of the susceptible potato cultivar to survive the transformation process was because too many cells accumulated virus and the effects were too debilitating. If this simple model is correct, then the approach of transforming plants with entire genomes has the potential for exploring the mechanism(s) by which cells can resist PLRV multiplication after the virus RNA genome arrives in the cell. This contrasts with the normal, and more complex, situation in which virus inocula must travel within the inoculated plant in order to reach the tissues in which resistance might be expressed.

This approach thus separates the infection process into two phases. The extra-cellular phase includes biological effects on the vector and on the host. In other words, it is a compound of the components of the delivery process. The intra-cellular phase concerns the establishment of the infection and the accumulation of the progeny virus. This novel dissection of the infection process could lead us to new insights into how plants naturally resist infection by PLRV.

Variation among aphid vectors of *Potato leafroll virus*

J.A.T. Woodford, B. Fenton & M.A. Mayo

Potato leafroll virus (PLRV) and its main vector, the peach-potato aphid, *Myzus persicae*, have been studied extensively at SCRI. The mechanisms involved in the transmission of PLRV are both scientifically interesting (transmission is a highly specialised process), and economically important because infection with PLRV is a serious problem for seed potato production. Current measures to prevent the spread of PLRV in Scotland involve the intensive use of insecticides to prevent the build-up of potential vector populations. Understanding more about epidemiological aspects of PLRV biology and, in particular, the transmission process of PLRV, will enable us to pinpoint those aphid populations responsible for spreading the virus. This is a prerequisite for developing novel, more environmentally sensitive control methods.

PLRV is mainly confined to phloem tissues of infected plants. Only aphid species that colonise potato plants are natural vectors because they must feed from the phloem to acquire the virus. Transmission involves the passage of ingested virus particles from the gut to the haemocoel and their subsequent transport through the accessory salivary glands and into saliva, where they can infect a new host. Most attention has been focused on properties of the virus particles that are recognised by aphid tissues. Several studies suggest that proteins on surfaces of virus particles play a key role in transmission. Early progress in this area was made with the discovery of poorly aphid-transmissible (PAT) isolates of PLRV that lack epitope(s) found on the surface of particles of highly transmissible (HAT) isolates. Work at SCRI has also suggested that changes in the amino acid sequence of the readthrough protein might account for alterations in efficiency of transmission. However, more recent transmission experiments with virus-like particles that lacked readthrough protein (*Ann. Rep. 1996/97*, 164), led us to revise this hypothesis, and to consider the role of vector components in transmission. Less is known about vector proteins involved in transmission, but clones of *M. persicae* that are inefficient vectors of PLRV would be a useful model for comparative experiments.

At least 10 aphid species colonise potato foliage and ingest phloem contents, but they do not all transmit PLRV efficiently. For example, the potato aphid, *Macrosiphum euphorbiae*, is usually the most numerous species in Scottish potato crops, but it transmits PLRV less efficiently than *M. persicae*. Moreover, in addition to differences in the efficiency with which PLRV is transmitted by aphid species, there are reports of differences in transmission efficiency for individual clones of *M. persicae*. Distinguishing such clones is difficult because they show remarkably little morphological or biochemical variation. However, the recent development of a DNA fingerprinting method enables us to characterise clones by variability in the lengths of the IGS regions between ribosomal genes (*Ann. Rep. 1997/98*, 126). We have therefore been able to compare the transmissibility of PLRV using distinct aphid clones, and to re-examine the transmission of PAT isolates of PLRV. Collaborative work with colleagues at the Institut National de la Recherche Agronomique (INRA) at Le Rheu in Brittany, France has enabled us to extend these comparisons by including a French PAT isolate of PLRV, as well as additional aphid clones.

Modern techniques for characterising aphid populations have led to surprising conclusions about the tax-

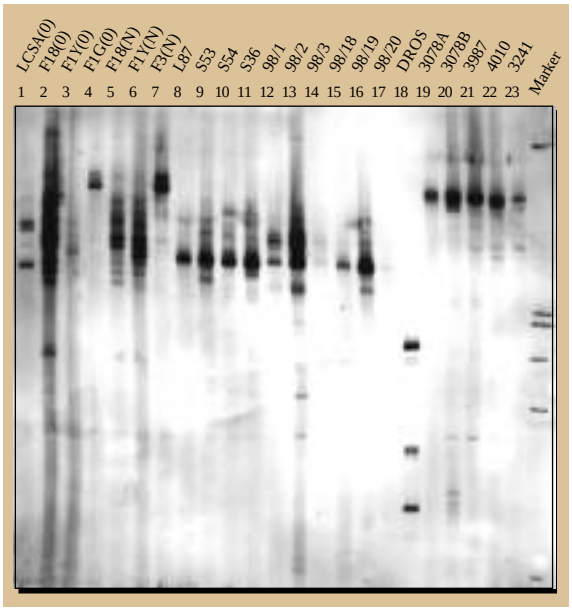

Figure 1 IGS fingerprinting of *Myzus* clones. Clones of *M. antirrhinii* from France (lanes 4,7) and England (lanes 19-23) have one or a few large bands, whereas clones of *M. persicae* have smaller band patterns that can be distinguished by number, position and intensity. LCSA (lane 1) shows a pattern that is similar by these criteria to those found in MP1S (lane 8) and other clones of from Scotland (lanes 9-17) and France (lanes 2,3,5,6).

onomy of *M. persicae*. Two distinct populations have been designated as separate species. A dark green form, separated as *Myzus antirrhinii*, which we have detected in Scottish potato crops (*Ann. Rep. 1997/98*, 126), was clearly distinguished from *M. persicae* clones, using the IGS fingerprinting method. *M. antirrhinii* clones showed much larger bands than were seen in clones of *M. persicae* (Fig.1). However, clone LCSA, a red aphid representative of the recently named species *Myzus nicotianae*, could not be distinguished from *M. persicae* by this method as it exhibited polymorphic bands of the same type as those found in *M. persicae*. To assess variation in transmission efficiency, we compared the standard *M. persicae* clone (Mp1S), which has been used for many years at SCRI, with other *M. persicae* clones, and several clones of *M. antirrhinii*, and with LCSA.

Aphids need to feed for several hours to acquire PLRV from infected plants, and the amount of virus

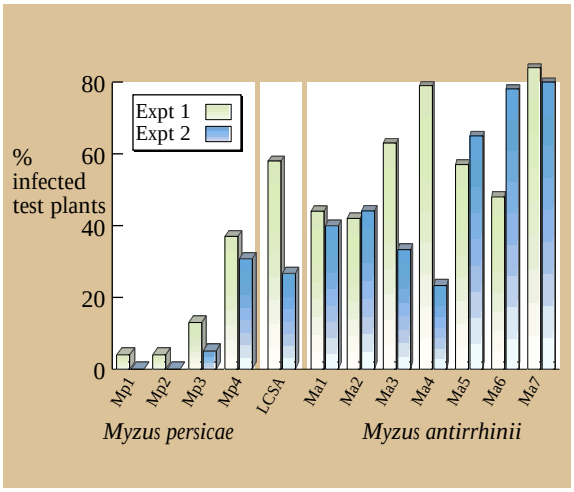

Figure 2 Aphid clonal variation in transmission efficiency after a 24h Acquisition Access Period (AAP) on *Physalis floridana* infected with PLRV-C.

acquired, and probability of transmission, increases with the duration of the acquisition feed. Long 'acquisition access periods' (AAP) on infected plants, and 'inoculation access periods' (IAP) on test plants are useful to find out if an aphid species can transmit. In previous studies with other species, we gave aphids AAP and IAP of 3-6 days, but we used shorter AAP to demonstrate variations in vector efficiency among *Myzus* clones. Fig. 2 shows the variation in the efficiency of 12 aphid clones in transmitting PLRV-C in two series of experiments, 9 months apart. Aphids were given a 24h AAP on excised leaves of PLRV-infected *Physalis floridana*, and then transferred, in groups of three, for an IAP of 3-5 days. There were consistent differences between clones. LCSA, and all of the *M. antirrhinii* clones, transmitted PLRV-C to 40% or more test plants, but only one of the *M. persicae*

Clone	% infected test plants	
	PLRV-V	PLRV-C
Mp1	8	53
LCSA	43	95

Figure 3 Clone LCSA transmits both PLRV-C and the 'poorly aphid-transmitted' isolate, PLRV-V, more efficiently than does Mp1S.

PLRV isolate	% transmission	
	Mp1S	LCSA
C 80 µg/ml	30	81
C 20 µg/ml	17	32
V 80 µg/ml	30	75
V 20 µg/ml	0	43

Figure 4 Clone LCSA transmits purified PLRV more efficiently than Mp1S in membrane feeding experiments, but the difference in transmissibility between the two PLRV isolates is detected only when virus is presented at lower concentration.

clones approached this efficiency. Transmission rates increased when the aphids were given a 48h AAP, but similar clonal differences were found. The poor efficiency of clone Mp1S in these tests was surprising because these aphids had been efficient vectors of PLRV-C in earlier experiments. If Mp1S had lost its ability to transmit PLRV efficiently, it would no longer be possible to discriminate between HAT and PAT isolates of PLRV with this clone.

In earlier tests when aphids had been given an AAP of 6 days on infected potato plants, Mp1S transmitted PLRV-V much less efficiently than PLRV-C. Repeating these tests more recently with Mp1S and LCSA, we still found that PLRV-V was transmitted less efficiently, but that transmission also depended on the aphid clone. LCSA transmitted both isolates more efficiently than did Mp1S, causing infection with PLRV-C in almost twice as many test plants, and five times as many with PLRV-V (Fig. 3). Moreover, tests in France showed that other aphid clones could also transmit PLRV-V more efficiently than did Mp1S, although never as efficiently as they transmitted HAT isolates. LCSA was also an efficient vector of the French isolate, 14.2, which could not be transmitted, or only poorly transmitted, by several clones of *M. persicae*, including Mp1S. These effects are not thought to result from lower concentrations of virus in plants infected with PAT isolates because their virus contents in ELISA tests were similar, but they could depend on differences in aphid feeding behaviour, or differences in the availability of the different isolates at aphid feeding sites. Indeed, differences in aphid feeding behaviour might well account for the large variations in the rates at which different clones transmitted PLRV-C (Fig. 2).

To examine these differences between virus isolate and aphid clone in more detail, and eliminate variations in virus content in leaves, aphids were fed purified virus through stretched Parafilm-M® membranes. In these tests, Mp1S and LCSA were confined for a 24h AAP on sachets of 20% sucrose containing PLRV-C or PLRV-V at two concentrations. Again, LCSA was a more efficient vector than Mp1S for both isolates, but transmission rates depended on virus concentration. It was only possible to distinguish the transmissibility of the isolates when they were presented at lower concentrations (Fig. 4). When aphids were fed through membranes on PLRV at
80mg/ml, both isolates were transmitted efficiently by LCSA, and moderately efficiently by Mp1S, but at

20mg/ml, Mp1S transmitted PLRV-C, but not PLRV-V, whereas LCSA transmitted both isolates equally efficiently.

These results show that the ability of aphid vectors to transmit PLRV depends not only on the structure of the virus coat protein, but also on biological factors contributed by the aphids themselves. There were large interspecific differences between aphids in vector competency for PLRV-C when sensitive assays were used. The tested clones of *M. antirhinii* came from geographically widespread sites, and comprised two distinct karyotypes, but all of them were more efficient vectors than was Mp1S. There were also intraspecific differences in transmission efficiency between the clones of *M. persicae.*

It is premature to conclude that qualitative or quantitative differences between recognition sites for PLRV could account for these differences in transmission rates. The feeding behaviour of the clones may differ, even when they are exposed to virus in sucrose diets. Current experiments are being made to compare feeding rates of efficient and inefficient vector clones during AAP on virus preparations. Mp1S is unable to reproduce sexually, and has been kept in parthenogenetic culture at SCRI since 1977. Interestingly, the two least efficient French clones of *M. persicae* had also been cultured at Le Rheu for many years. Although this may indicate some long term effect of laboratory culture on transmission efficiency, recent evidence from Scotland suggests that the low vector competency of the Mp1S genotype is not confined to the laboratory culture. In 1996, we found that the IGS pattern characterising Mp1S was found in some 30% of *M. persicae* samples from eastern Scotland. In 1998, almost all clones of *M. persicae* that were derived from migrant alatae colonising potatoes and brassicas at Invergowrie gave an identical IGS fingerprint to that of Mp1S (Fig. 1). The vector competency of three of these 1998 clones that have been tested is no greater than that of the standard Mp1S. If this result proves to be typical for other samples of the Mp1S genotype, it could have important implications for the epidemiology of PLRV in Scotland. Aphids with this IGS fingerprint have been found in Scottish samples over several years and, at present, it appears to be the predominant clone. Amongst UK populations of *M. persicae*, Mp1S is unusual in being susceptible to insecticides. Careful monitoring should ensure the early detection of any changes in the clonal composition of *M. persicae* or closely related species on seed potato crops that could herald the arrival of more efficient vectors and increase the risk of spread of PLRV.

Acknowledgement

We thank Drs Danièle Bourdin and Yvon Robert for providing aphid clones and facilities to work at INRA, Le Rheu. Part of this work was funded by the British Council, Paris under the Franco-British Joint Research Programme (Alliance).

The novel mitochondrial genome architecture of the potato cyst nematode *Globodera pallida*

M.R. Armstrong, V.C. Blok, M.S. Phillips & D.L. Trudgill

Mitochondria are oval-shaped organelles present in the cytoplasm of all eukaryotic cells. They are the site of the essential metabolic process of oxidative phosphorylation. Each mitochondrion contains a small chromosome which encodes products necessary for mitochondrial function. Typically, metazoan mitochondrial DNAs (mtDNAs) are single circular molecules between 13.5 and 30 kb in length that encode 22 transfer RNAs (tRNAs), two ribosomal RNAs (rRNAs) and 12 or 13 proteins involved in oxidative phosphorylation and electron transport. The inability to detect a mtDNA of sufficient length to encode the typical metazoan mitochondrial gene complement, in populations of the Potato Cyst Nematode, *Globodera pallida*, is a unique departure from conventional models of metazoan mtDNA structure. In *G. pallida*, at least six small circular mtDNA (scmtDNA) molecules are typically found, in populations from a wide variety of locations. These scmtDNA molecules vary in size from 6.3 to 9.5 kb (Fig. 1). No evidence for a larger 'wild type' molecule has been found using PCR, Southern blotting or electron microscopy (Fig. 2), although the presence of such a molecule at low levels

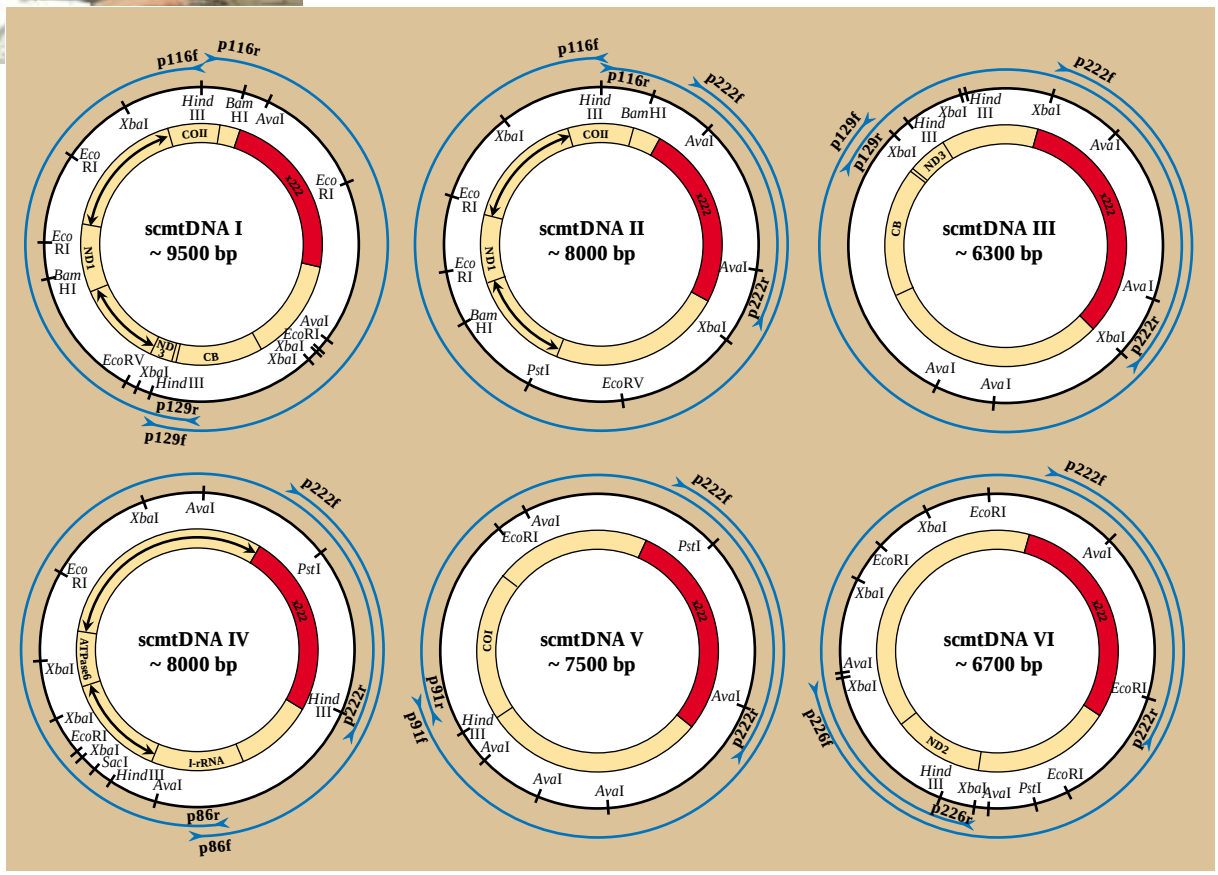

Figure 1 Restriction maps of the six scmtDNAs referred to in the text. The common ~2 kb non-coding region present on each scmtDNA is coloured red. Preliminary data from hybridisation studies regarding the distribution of mitochondrial gene coding sequences among the scmtDNAs is indicated. Note that scmtDNA IV alone contains rRNA sequences.

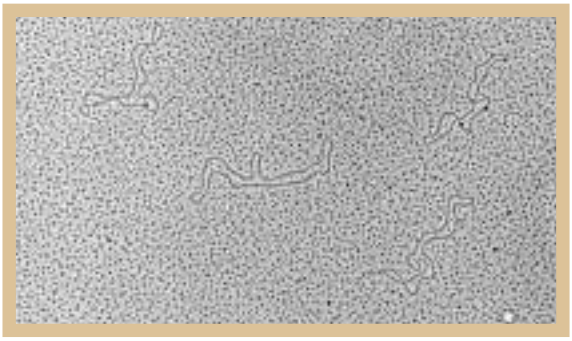

Figure 2 Electron micrograph of *G. pallida* scmtDNA molecules. The four molecules shown were estimated to range in size from 7.4 - 10 kb. No molecule longer than 10.5 kb was identified from over 150 such measurements.

cannot be ruled out. Each scmtDNA has been demonstrated to contain mitochondrial gene coding sequences and a ~ 2 kb non-coding region, which is common to all scmtDNAs. Preliminary investigations have suggested that all 12 mitochondrial protein coding genes found on nematode mtDNAs are distributed amongst the aforementioned scmtDNAs. In particular, rRNA genes (which are necessary for the translation of mitochondrial proteins) are found on only one scmtDNA. This raises the possibility that the components of the mtDNA operate in concert to encode the products necessary for mitochondrial function.

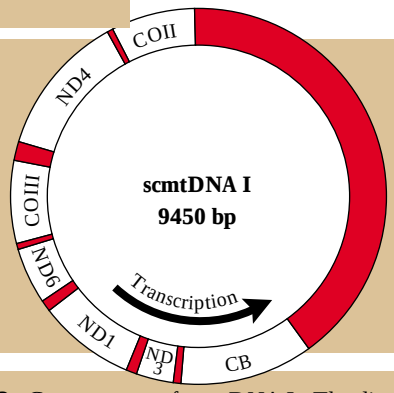

Figure 3 Gene content of scmtDNA I. The direction of transcription of the seven mitochondrial genes is indicated by the arrow. The 3545 bp non-coding region, containing a sequence similar to the non-coding regions present on all other scmtDNAs, is coloured red as are the six short intergenic sequences.

The complete sequence of scmtDNA I has been obtained. Analysis of this sequence indicates that it encodes seven full length mitochondrial proteins. Other than these genes, no regions of this molecule were identified that could encode further mitochondrial genes. Significantly, no rRNA genes were identified. These observations provide support for the notion that scmtDNAs in general are functional, while also confirming that scmtDNA I in particular would not be functional in isolation. The gene content of scmtDNA I is presented in Figure 3.

The presence of certain scmtDNAs was found to be unvarying in European and South American populations. For example, scmtDNA IV is invariably found. This might be expected given the presence of rRNAs on this scmtDNA. However, some scmtDNAs were found to vary enormously in their abundance . For example, in the majority of European populations, scmtDNAs II and III are the dominant mitotypes, while scmtDNA I is not detectable by Southern hybridisation. However, in an unusually virulent population (Luffness), scmtDNA I is the dominant mitotype ,while scmtDNAs II and III are undetectable (Fig. 4). This may suggest that the multipartite state affords a degree of flexibility in genome organisation as scmtDNAs I, II and III each contain some coding sequences in common (Fig. 1).

Potato cyst nematodes, particularly *G. pallida* in the U.K., are important agricultural pests. European populations exhibit considerable variation in virulence, but the lack of qualitative sources of resistance has prevented the establishment of useful phenotypic classifications among the majority of populations. Analysis of scmtDNA sequence variation has provided new insights as to the relationship between European *G. pallida* populations and their indigenous South American

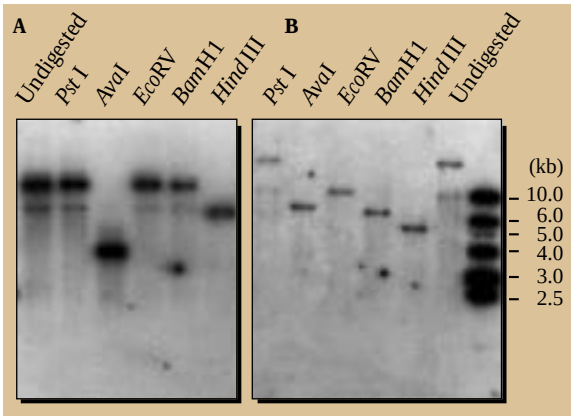

Figure 4 Southern blot of total genomic DNA from population Gourdie (Panel A) and Luffness (Panel B) hybridised with a probe specific to both scmtDNAs I and III. The restriction fragments detected in Panel A (Gourdie) are consistent with having originated from scmtDNA III. The restriction fragments detected in Panel B (Luffness) are consistent with having originated from scmtDNA I.

ancestors. Figure 5 shows a schematic representation of sequence variants of scmtDNAs I-IV found among five key populations, two South American and three European. South American populations contain a more diverse population of scmtDNA sequence variants than European populations. For example, only one of the four scmtDNAs typically found in European populations was detected in South American population P5A and this was a highly diverged sequence variant of scmtDNA IV. This suggests that this population has historically experienced low levels of gene flow with populations like P4A. Figure 5 suggests that population P4A also contains a diverse range of scmtDNA molecules. All the scmtDNAs found in the British populations Gourdie and Luffness are subsets of those present in population P4A. A third British population, Pa1, contains some novel scmtDNA sequence variants, suggesting it may have been derived from a distinct gene pool to that which gave rise to populations like Gourdie and Luffness.

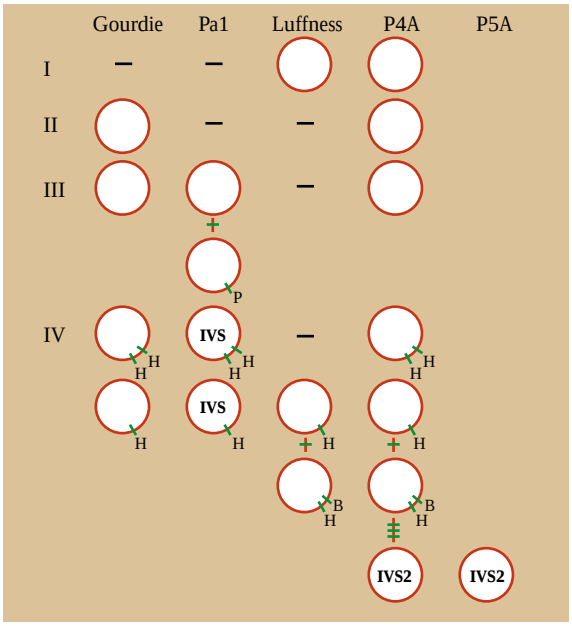

Figure 5 Schematic representation of sequence variants of scmtDNAs I-IV found in populations Gourdie (U.K.), Luffness (U.K.), Pa1 (U.K.), P4A (South America) and P5A (South America).

Implications The novel model of mtDNA structure presented above raises a number of fundamental questions: How is scmtDNA replication in *G. pallida* co-ordinated and gene expression regulated? The respiratory complexes that make up the electron transport chain are encoded as subunits present on both the nuclear and mitochondrial genomes. Increases in mitochondrial biogenesis therefore require co-ordinated increases in gene expression in both the nuclear and mitochondrial compartments. A multipartite mtDNA would impose an additional layer of complexity on this system, as the expression of individual parts of the mtDNA would presumably also have to be co-ordinated. In this regard, it may be significant that each scmtDNA contains an unusually lengthy non-coding region. With the exception of repetitive elements, metazoan mtDNA non-coding sequences are generally far shorter than those found on *G. pallida* scmtDNAs.

How is the essential gene complement of a multipartite genome maintained during both somatic and germ line cell divisions? In contrast with the replication of the nuclear genome, mtDNA replication is unpredictable. While nuclear DNA replication invariably results in the generation of a precise copy of each chromosome, each of the many mtDNA molecules present in a cell is not faithfully copied. As a result, cells containing mixed populations of mtDNA molecules (heteroplasmic cells) can give rise to cells or gametes that contain only one type of mtDNA molecule. In conventional systems this process can be very rapid, with the parental mtDNA genotype being replaced in one generation. With a multipartite mtDNA, it would be necessary to counteract this effect to ensure the viability of progeny.

Other questions concern the mechanism by which an apparently fragmented mtDNA arose and the implications for mtDNA genome evolution. For example, do the components of the *G. pallida* mtDNA recombine and what are the processes that determine the frequency a particular scmtDNA is found at in a population? It is also unclear how widespread this phenomenon is within the Nematoda.

Progress towards Integrated Crop Management (ICM) for European raspberry production

S.C. Gordon, J.A.T. Woodford, B. Williamson, A.N.E. Birch & A.T. Jones

Raspberries are an important, high-value, horticultural crop grown without subsidy in many northern European countries. There is also an increased production in more southern countries such as France, Greece, Italy, Portugal, Spain, and Switzerland. Total production in Europe in 1997 was estimated to be *c.*240,000 tonnes[1]. Whilst most raspberries are grown for the fresh market, a high proportion of the Eastern European, French and Scottish raspberry industries, is used for processing.

Arthropod pests and diseases of cane fruits may cause both direct and indirect damage and loss. At present, methods to protect crops against such effects rely largely on the application of broad-spectrum insecticides and fungicides. The number and spectrum of active ingredients in these products approved for use on cane fruit crops is declining rapidly across Europe, due to the high cost of registration for use in this minor crop. In the very near future, growers will need to find alternative methods to manage pests and diseases, using fewer applications of a very limited number of products. At the same time, consumers and processors are encouraging growers to continue to produce high quality fruit with the minimum amount of pesticides. These demands will inevitably lead to changes in crop management.

Direct damage to berry fruits through yield loss or fruit blemish caused by arthropods, includes that caused by raspberry beetle (*Byturus tomentosus*), clay-coloured weevil (*Otiorhynchus singularis*), raspberry moth (*Lampronia rubiella*) and two-spotted spider mite (*Tetranychus urticae*). Direct damage is also caused by grey mould fungus (*Botrytis cinerea*). Indirect damage by the aphids, *Amphorophora idaei* (*Am. idaei*) and *Aphis idaei* (*A. idaei*) and the leafhopper, *Macropsis fuscula*, is caused by the transmission of plant viruses and the phytoplasma agent of rubus stunt respectively (Table 1). Additionally, the wounding caused by the raspberry cane midge (*Resseliella theobaldi*) predisposes plants to fungal infection[2]. Furthermore, if insects are not adequately controlled on fruiting plants before and at harvest, the risk of contamination of the harvested fruit with these organisms is greatly increased. The most common arthropods causing rejection of fresh and processed fruit are aphids, earwigs, larvae of beneficial predators, and parasites.

Insecticide and fungicide usage in raspberry Contact organophosphorus-based pesticides account for about 70% of the insecticide and acaricide use in raspberry crops in the UK. They are used mainly to control raspberry beetle and raspberry cane midge. Systemic organophosphorus-based products, targeted primarily against aphids, currently account for less than 5% of insecticide usage and this is due to the widespread and increased cultivation of aphid-resistant raspberry cultivars. However, the need for aphicides may increase as the incidence of resistance-breaking raspberry aphid biotypes become more widespread. The use of acaricides is also declining as growers turn to biological control of two-spotted spider mite. Broad-spectrum fungicides, such as dichofluanid that is used widely during blossom to control grey mould, have additional benefits by controlling other fungal diseases, such as cane

spot (*Elsinoe veneta*), spur blight (*Didymella applanata*) and stamen blight (*Hapalosphaeria deformans*). Although more target specific fungicides are available or being developed, their use may lead to damaging outbreaks of the fungi currently controlled by the broad-spectrum products. Changes in husbandry practice, such as protected cultivation or machine harvesting, have given rise to new problems and they will result in the on-going need for fungicides.

Monitoring, scouting and forecasting Many of the components of an ICM programme for raspberries are in place but real progress will depend on their implementation at the farm level. This will inevitably place a greater management burden on fruit growers and technical support staff, as they will have to collect and assimilate more information on the health of the crop and make more decisions on its treatments. The information that they need to gather will be based on regular crop walks during daylight hours and after dark to check for pests and pathogens, the use of insect traps and the knowledge of control thresholds for spray strategies, and the use of models to predict the onset of pest and disease attack. Useful information about the health status of canes can be gained by routine sampling during winter of, for example, midge blight and cane spot.

The raspberry cane midge oviposition model is a good example of a system that successfully predicts the onset of insect attack in the spring. This permits growers to assess the risk of damage to their crop by assessing midge levels in the previous year and to decide, if and when, to apply insecticides to manage first generation midge numbers. Numbers of the raspberry beetle, the most important flower and fruit pest in northern Europe, can be assessed by examination of flower buds in the spring. Recently, white sticky traps (Rebell® bianco), developed in Switzerland, have been shown to trap adult raspberry beetles. They are currently being assessed to develop a spray threshold in various European countries, as part of the 'Reduced Application of Chemicals in European Raspberry production' (RACER) project (Fig. 1). Successful development of ICM will require a planned, multi-national approach, with transfer of existing research to new areas and its adaptation to the local environmental conditions. Although monitoring and forecasting pest and disease incidence is an important part of ICM systems, they still rely on having the chemicals or strategies to control the pest or pathogen. However, many of the former are under threat of being withdrawn from use. There is now an urgent need for the registration and release of newer, more environmentally benign, pesticides that are effective against common raspberry pests and diseases.

Figure 1 The RACER project was conceived by members of the European raspberry producers and processing industry to help them to produce high quality fruit in an environmentally acceptable manner, meeting the aspirations of the consumers, supermarkets and processors. http://www.scri.sari.ac.uk/Racer/

Plant resistance Plant resistance, provided it is durable, remains one of the most efficient, benign and cost-effective means of pest and disease control. Several examples of such resistance occur in raspberry. One of the best documented and utilised is resistance to the large raspberry aphid, *Amphorophora idaei* (*Am. idaei*). The major importance of this aphid is as a vector of at least four different viruses of raspberry (Table 1). These viruses, either alone or in combination, cause serious losses in plant growth and vigour, and

	Common name†	Latin Name	Type of Damage	Distribution in Europe	Importance
Pests					
	Large Raspberry Aphid	*Amphorophora idaei*	Virus Vector/Foliage	Widespread/Northern	*****
	Small Raspberry Aphid	*Aphis idaei*	Virus Vector/Foliage	Widespread/Southern	**
	Rubus leafhoppers	*Macropsis* spp.	MLO Vector	Localised	*
	Common Green Capsid	*Lygocoris pabulinus*	Foliage	Localised	*
	European Tarnished Bug	*Lygus rugulipennis*	Foliage/Flowers	Widespread/Northern	*
	Raspberry Beetle	*Byturus tomentosus*	Flowers/Fruit/Contaminant	Widespread/Throughout	*****
	Clay-coloured Weevil	*0tiorhynchus singularis*	Buds/Foliage	Localised/Northern	***
	Strawberry Blossom Weevil	*Anthonomus rubi*	Buds/Flowers	Localised/Southern	**
	Raspberry Cane Midge	*Resseliella theobaldi*	Canes (Midge Blight)	Widespread/Throughout	*****
	Raspberry Moth	*Lampronia rubiella*	Buds	Localised/Northern	***
	Double Dart Moth	*Graphiphora augur*	Buds	Localised/Scotland	***
	Two-Spotted Spider Mite	*Tetranychus urticae*	Foliage	Widespread	****
	Raspberry Leaf and Bud Mite	*Phyllocoptes gracilis*	Foliage	Widespread but sporadic	***
Diseases					
	Fungal diseases				
	Grey Mould	*Botrytis cinerea*	Canes/Fruit	Widespread	*****
	Cane Blight	*Leptosphaeria coniothyrium*	Canes	Widespread	***
	Midge Blight (Disease complex)	*R. theobaldi* + *Phoma/Fusarium*	Canes	Widespread	*****
	Cane Spot	*Elsinoe veneta*	Canes/Leaves/Fruit	Widespread	**
	Raspberry yellow rust	*Phragmidium rubi-idaei*	Leaves	Widespread	**
	Root-rot	*Phytophthora fragariae* var *rubi*	Roots	Widespread	*****
	Am. idaei-borne viruses				
	Symptomless decline	*Black raspberry necrosis virus*	Latent	Widespread?	*
	Leaf spot mosaic/decline	*Raspberry leaf mottle virus*	Foliage/Vigour	Widespread?	*****
	Leaf spot mosaic/decline	*Raspberry leaf spot virus*	Foliage/Vigour	Widespread?	*****
	Yellow mosaic	*Rubus yellow net virus* (RYNV)	Foliage	Widespread?	*
	Veinbanding mosaic/decline	RYNV + other viruses	Foliage/Vigour	Widespread?	****
	Aphis ideai-bourne viruses				
	Vein chlorosis	*Raspberry vein chlorosis virus*	Foliage	Widespread	**
	Pollen-borne virus				
	Bushy dwarf/Yellows	*Raspberry bushy dwarf virus*	Foliage/Fruit	Widespread	**?
	Nematode-borne				
	Yellow dwarf/decline	*Arabis mosaic virus*	Foliage/Vigour	Localised/ Northern	***
	Scottish leaf curl/decline	*Raspberry ringspot virus*	Foliage/Vigour	Localised/ Northern	***
	Decline	*Strawberry latent ringspot virus*	Foliage/Vigour	Localised/ Northern	**
	Ringspot/decline	*Tomato black ring virus*	Foliage/Vigour	Localised/ Northern	***
	Leafhopper-borne agent				
	Rubus stunt	Phytoplasma	Foliage/flowers	Continental Europe	***

MLO = Mycoplasma like organism.
† Common name in the UK, may vary elsewhere in Europe

***** = Of major importance, causing severe loss/damage if not controlled;
**** = Important, severe loss/damage;
*** = Locally important causing moderate/severe damage;
** = Locally important causing slight/moderate damage;
* = Minor, cosmetic damage

Table 1 Major pests and diseases of raspberry in Europe: their damage, distribution and importance.

fruit yield and quality[3]. Gene A_1 and uncharacterised minor genes from red raspberry confer effective resistance to some common biotypes of this aphid, and gene A_{10} from American black raspberry (*R. occidentalis*) is effective against all described biotypes. Over the last 20 years in Britain, control of this aphid vector by these resistance genes has also given very effective control of the four viruses it transmits, without the need for aphicides[3]. However, very recently, *Am. idaei* has been found colonising gene A_{10}-containing cultivars in Britain, suggesting that new A_{10}-breaking biotypes of this aphid have developed. No immunity or resistance has been found in *Rubus* germplasm to any of the four viruses transmitted by *Am. idaei*, so that the control of the spread and effects of these viruses continues to depend on effective control of this aphid vector. In the absence of resistance genes against the new *Am. idaei* biotypes, alternative control strategies for this common aphid need to be devised urgently.

The small raspberry aphid, *A. idaei*, common on raspberry throughout continental Europe, seems to have increased in incidence in more northern latitudes. It is generally regarded as of low significance as a pest and derives its importance only as the known vector of raspberry vein chlorosis virus (RVCV). This virus, that affects plant growth and fruit quality, has increased in incidence in crops in recent years. No sources of resistance to *A. idaei* have been identified in raspberry but immunity to RVCV in some North American *R. idaeus* var. *strigosus* cultivars has been identified. Work at SCRI has shown that immunity or very strong resistance to RVCV can be introduced fairly readily into raspberry from these sources of immunity[4], offering an effective means of controlling this virus.

The pollen-borne, raspberry bushy dwarf virus (RBDV) is common in raspberry world-wide. In sensitive cultivars it causes yellows disease and/or crumbly fruit that greatly affects fruit quality[5]. In combination with aphid-borne viruses, it can cause greatly decreased vigour and productivity (bushy dwarf disease). A single dominant gene *Bu*, present in several cultivars, provides immunity to common (S) strains of RBDV, and this gene has been effective against RBDV infection in the field world-wide for more than 50 years. However, the virus is increasing in prevalence in many countries due to the increased planting of cultivars lacking this gene and, in some localities, to the occurrence of strains of this virus, termed resistance-breaking (RB), that can infect cultivars immune to S strains. The occurrence of such RB strains is of serious concern because plant resistance is the only means of controlling this virus in crops. Because of this impasse to the control of RB strains, work is underway at SCRI to produce, evaluate and assess the risks of transgenic resistance to RBDV, using various constructs of different regions of the viral genome.

Currently, control of the four nematode-borne viruses that affect raspberry in Europe, depends on the use of soil fumigants to kill the nematode vectors and the application of herbicides to remove virus sources. Whilst this has been very effective for viruses transmitted by *Longidorus* nematodes (raspberry ringspot and tomato black ring), it has been much less so for those viruses transmitted by *Xiphinema* nematodes (arabis mosaic and strawberry latent ringspot). This is because vectors retain the latter viruses for very long periods. Genes for immunity to one or more of the four viruses have been identified in raspberry but they are not effective against all strains of the viruses involved, making their deployment of little value commercially. With the impending withdrawal of some widely used soil fumigants from commerce, alternative control strategies for these viruses are required. One possibility being explored by SCRI is the use of virus sequences as transgenes in raspberry and preliminary experiments in a *Nicotiana* model have shown the efficacy of this approach. It remains to be seen if this approach is effective in raspberry and what risks may be associated with its use.

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Plants, soils and environment

John W. Crawford

Work within the Plants, Soils and Environment theme is concerned with improved understanding of the ecological, physical and biological processes which impact on the management of agricultural and semi-natural systems. These processes operate across a vast range in scale from the molecular to the regional. They do not operate in isolation but are strongly interactive, and the greatest challenge in our research is to develop our understanding to account for the role of these interactions in the system-scale behaviour. The work is therefore integrative and multidisciplinary, and interfaces with most of the thematic programmes at the Institute. Our applied focus is underpinned by a strong fundamental and strategic science base.

Agriculture is facing its most important challenges as the millennium comes to a close. On a global scale, population is expected to increase by 64% over the next 20 years and if global nutrition is to be brought to a more equitable state, this will be accompanied by a substantial increase in the demand for food. At the same time, a reduction in the natural fertility of soil is causing an estimated 17% decrease in productivity and, although pesticide applications have increased 10-fold since 1945, crop losses have doubled as we lose the selection battle against the development of natural resistance. Against the background of these challenges, it is being realised that intensification, and the associated high inputs required by current agricultural production, is being realised at an unsustainable cost to the environment. For example, agriculture is now the main diffuse-source polluter of water. Biotechnology offers some solutions to these problems with the potential for engineering resistance and increased nutritional quality, and restoring soil fertil-

ity through remediation and structural regeneration. However, many of these technologies are in their infancy and some have their own potential problems. Durability of engineered resistance, the acceptability of genetic engineering, and concern over environmental risk are notable examples. Simply reducing inputs and putting more land into production is not the answer on its own either, since it is abundantly clear that increased extensification of agriculture has been, and would continue to be, a significant factor in the destruction of natural habitats. Therefore, as we strive for more sustainable food production, we have to recognise that no single solution is going to be the right one, and that it is likely that most progress will be made by integrating different approaches to achieve the best compromise. This requires that we understand not only a large number of different processes, but also the nature of their interactions and the consequences for the large-scale behaviour of the system.

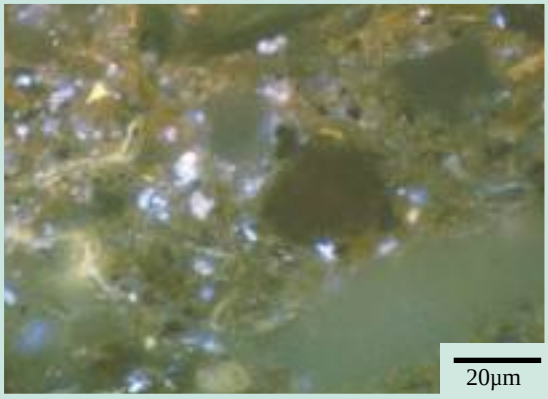

Figure 1 Thin section of soil (brown) showing bacterial colonies preserved *in situ* (fluorescent blue).

The soil system is simultaneously our most valuable and abused natural resource. Its physical architecture conducts water flow to the groundwater reservoir, while retaining sufficient quantities in the upper horizons to mediate aeration and to sustain plant, soil faunal and microbial life. Within this structurally complex habitat resides an enormously diverse microbial community. A single gram of arable soil contains more biological diversity than the entire mammalian kingdom and concurrently performs millions of complex biochemical transformations that recycle dead organic material into a form once more available for primary production. If sustainability is to be realised, this natural fertility will have to be exploited through improved management that is beyond the grasp of current understanding. We are developing theoretical and experimental approaches to account fully for the interactions between soil structure and biological functioning. As a first step, it is essential that we have techniques that allow us to directly visualise the soil physical architecture with the microbial community *in situ*. During the year, we have perfected methods for fixing the soil in hard transparent resin that are sufficiently non-intrusive that delicate biological material such as bacterial cells and fungi is perfectly preserved. This resin-impregnated soil can be cut into thin wafers and polished to a few thousandths of a millimetre in thickness, to provide us with thin sections that can be viewed down a microscope (Fig. 1). By employing a fluorescent stain of the microbes, the optical contrast between the physical architecture and the soil can be enhanced. Using digital image capture, we can process the images and analyse the spatial structures that exist both in the architecture of the soil and the distribution of the microbes. Thus, for the first time, detailed statistics relating to the relative arrangement of soil and microbes can be obtained. This precise quantification is required in order that the information can be used in mathematical theories that are being developed in tandem with our experiments on the functioning of the soil system. Such an approach is being followed in a project funded during 1998 under the Government's DTI Biological Treatment of Soil and Water LINK scheme. In the largest award made under the scheme, the DTI has commissioned the Soil Plant Dynamics Unit to work with one of the world's

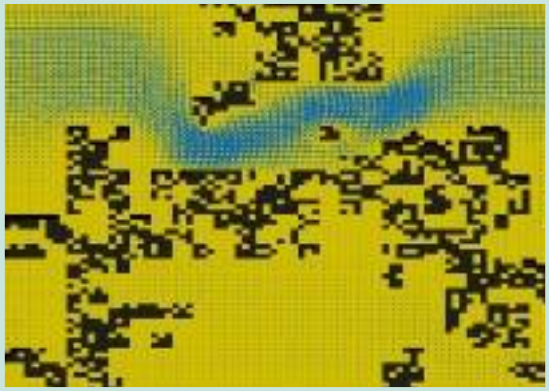

Figure 2 Computational fluid dynamics models are used to understand the impact of structure on flow paths and solute residence times.

largest agrochemical companies, Rhone Poulenc, and an environmental consultancy company, Quantisci Ltd., to study the fate of organic chemicals in soil. This project acknowledges the fact that the major uncertainties in predicting the fate of reactive compounds in soil reside in the link between pore-scale structure and the microbial activity that degrades the compounds. Using our recently developed imaging techniques, we will link the quantitative description of the soil system to models of transport with the aim of relating lab-based measurement to field-scale risk management. In a related 5-year project, SOAEFD have funded the Unit to lead an interdisciplinary research team combining groups from MLURI and Aberdeen University to study the fate of environmentally-important chemicals in two contrasting catchments: an upland nutrient-poor system, and a lowland arable system. Pore-scale models of surface reaction chemistry have been developed that demonstrate the significance of physical structure for adsorption-desorption processes, and are already providing clues to how the enormous complexity may be understood in terms of simple scaling relations between space and time. Computational fluid dynamic models of pore-scale solute flow are also being derived to understand how the structure of soil mediates the delivery of chemicals to the soil surface (Fig. 2). Our new approaches are providing insights into how the structure of porous media is related to the properties of reactive flows, with ramifications that go far beyond the current application. These pore-scale models are being linked to catchment scale models of solute flow to understand the role of heterogeneity in chemical transport at different scales (Fig. 3). In combination with the theoretical developments, a parallel programme of extensive field and laboratory studies of scaling effects on hydraulic and chemical properties is being carried out. A catchment-scale arrangement of sample points has been designed and implemented to monitor the spatio-temporal scaling properties of the physical and chemical characteristics of the catchments. This is being related to underlying, topographical, geological and soil property information that is simultaneously being gathered and is beginning to reveal simple patterns that underlie the complex behaviour of the catchments. Perhaps the most challenging aspect of our work on reactive transport in porous media is to account for the impact of unsaturated conditions – the most common state of soil where not all the pore space is filled with water. In a project funded by SOAEFD, we have been developing experiments and theory to understand the consequences of soil structure for the ability of the soil to retain moisture against suction, such as is applied by plant roots, and to understand the effect of unsaturated conditions on the connectivity of the water films. The latter property is crucial for understanding transport of oxygen, solutes, nutrients and/or microbes in unsaturated soil. By comparing our theoretical models with the results of retention measurements and quantification of structure from thin sections, we found our predictions to be accurate to greater than 5%. This confirmation of our hypotheses has revealed a fascinating scaling relation between the connectivity of water films and the applied suction. This relation leads to an important simplification in the theory and implicates a single parameter as the governing factor in determining the rate at which

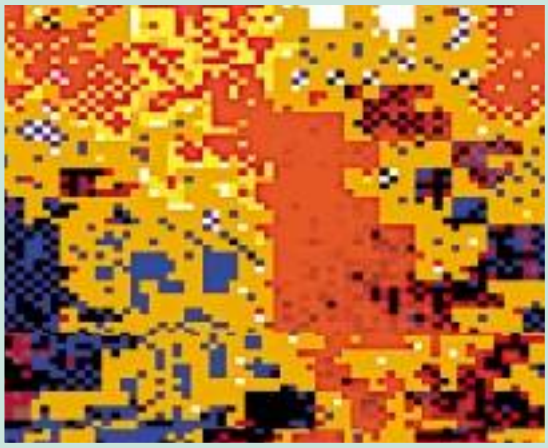

Figure 3 Simulation of chemically-reactive solute flow in a fractal soil structure (brown). Red to blue denotes high to low concentration respectively.

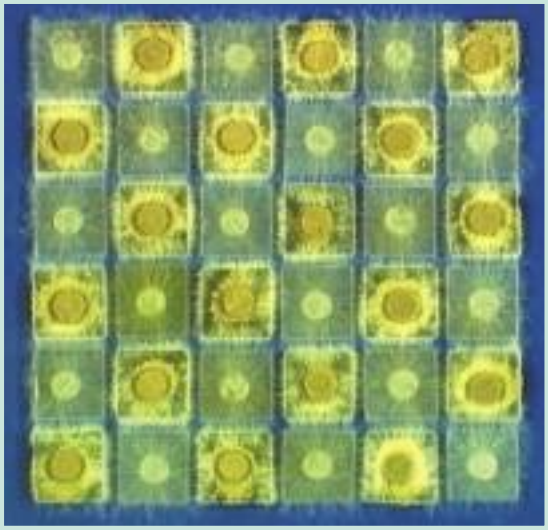

Figure 4 Tessellated agar tile system to study emergent behaviour in fungal communities.

air enters structured porous media under pressure. A fuller account of the approaches to the study of the fate of environmental compounds is provided in the Report of the Soil Plant Dynamics Unit elsewhere in this volume.

Much of our soil-related research concerns the functioning of the soil microbial community, and in both our research on microbial and plant systems, the relation between community structure and spatio-temporal dynamics is of central concern. This link is being studied in the context of soil fungi because of their important role in natural fertility, soil structure generation, soil-borne diseases and their potential for bioremediation (Fig. 4). Being non-determinate, they also belong to a wide class of organisms whose community dynamics has not been studied in detail before. In collaboration with the University of Abertay Dundee, we have been examining the dynamics of a fungal microcosm using a combination of experimental and theoretical techniques to focus on the relative importance of small and large-scale context for the behaviour of the community. We found the hypothesis that the large-scale dynamics is a consequence of independent interactions between species in a local neighbourhood can be excluded at the 5% significance level. Instead, the system exhibits emergent behaviour whereby the outcome of interactions at the individual species level depends on community-scale features. Thus, it is unlikely that experiments performed out of the community context will provide useful information on the dynamics. New types of analyses and experiments are being developed to study these systems at the community level.

One of the most frustrating facts in relation to the microbial community in soil is that it is almost impossible to measure its diversity directly. As far as is known, only about 1% of the microbial community in soil can be extracted, grown in culture and measured, while the other 99% do not grow outside the soil system. The SOAEFD-funded MICRONET project entered its second phase in 1998 and concerns the connection between microbial community structure and function, and its relation to vegetation cover and land management in low-input upland systems. We have pioneered techniques for broad-scale analysis of community structure and these have been applied to understand how the genetic structure of communities differ at separate locations. One of the key hypotheses was that there might be a strong relation between the plant species and the microbial community that develops in the soil around them, at least in systems under the same management regime. This might arise from known effects that plants have on the nutritional and physical soil environment, and because the nature of that influence is plant species and land-management dependent. Surprisingly, however, we have discovered that adjacent communities under single-species swards are as genetically distinct as those separated by hundreds of kilometres. We could also find no significant effect of plant species. Therefore, either the soil environment is extremely variable despite the above- ground cover and hides any existing signal, or there simply is no relation between above and below-ground diversity. These questions are being pursued in Phase 2 of MICRONET using new molecular tools that provide a more detailed description of the part of the community most likely to be influenced by above-ground plant cover. Techniques are following on but narrowing in scope to focus on particular functional groups and we are developing novel molecular probes for nitrite reductase activity to characterise the denitrifying fraction of the community involved in denitrification. We have also secured two further grants under the NERC soil biodiversity initiative that aim to explore further the relation between the diversity of the microbial community and the soil environment. These projects will be carried out in collaboration with the Universities of Aberdeen and Stirling.

An additional link between above and below-ground processes is provided by our research on the dynamics of weed seedbanks. This work, carried out with substantial funding from MAFF and in collaboration

with ADAS, examines the effect of various levels of management intensity on the changes in diversity of weed seeds in the soil. Management options included crop rotation and herbicide, and, although there was some interaction between these, it was the latter that had the most significant impact on the seedbank. In general, seedbanks declined in total number as herbicide applications were increased. However, an interesting relation between species number and total seedbank was discovered, demonstrating the fact that increased weed species diversity is accompanied by an exponential increase in seedbank levels. Thus, under high herbicide inputs, very few weed species remained in the seedbank, but they coexisted in similar low abundance. Under low inputs, there were more species and some of these existed in very high abundance.

Our studies on vegetation systems continue the theme of exploring the role of individual behaviour in the dynamics of communities. The 5-year SOAEFD-funded Vegetation Dynamics programme is coordinated within the Plants, Soils and Environment Theme at SCRI and is a collaboration between SCRI, SAC and the MLURI. The overall goal is to understand the species composition in an upland grazed system, and we have responsibility for the theoretical aspects. A surprising finding of the work so far is the remarkable diversity that exists in what was previously thought to be a largely clonally-propagated system. The community is therefore rather well described as comprising distinct individuals. Parallel theoretical approaches are being used to synthesise this otherwise overwhelming complexity and to understand how such diversity can coexist. Results of the analyses so far indicate that communities of randomly assembled individuals cannot coexist in general and that the dynamical state and the stability of that state arise from interdependencies between the functional traits of individuals.

An important aspect of our studies on plant systems is the possible impact of genetically-modified organisms (GMOs) on the natural and semi-natural environment. Work is currently underway on a MAFF-funded project to study the regional-scale movement of oilseed rape pollen and to understand the role of regional-scale context on the rate of geneflow in the environment. Fields of oilseed rape were mapped across Tayside and bait plants and pollen traps were established to measure the spatial distribution of pollen and its viability. A theoretical model of pollen transport was also developed to aid in the interpretation of the results. We found that the measured distribution of pollen and therefore the potential rate of geneflow from agricultural fields into the environment is determined by the regional scale distribution of fields. The apparent rate of decline in pollen concentration from the nearest oilseed rape field is also determined by the regional context. This confirms our earlier conclusions that estimates of geneflow obtained from small isolated plot experiments tend to underestimate the characteristic distance over which geneflow can occur in a more realistic context.

Environmental flows

I.M. Young, A.G. Bengough, J. Chessell, J.W. Crawford, L. Deeks, X. Zhang

Background Flows of water, solutes and gases are fundamental to the major biogeochemical cycles that sustain terrestrial life. An important, yet poorly understood, component of these flows are those within the top 2 metres of soil covering the land surface. This edaphic zone is not only crucial for plant growth, but is highly relevant to the many man-made environmental concerns, such as application of agrochemicals and the disposal of waste chemicals and sewage sludge.

Central to our approach is the hypothesis that the spatial heterogeneity of soil architecture influences the flow and activity of solutes, and organisms, within a soil profile. We also recognise implicitly that the size-scale within which flow mechanisms operate, impact directly on flow characteristics. For example, at the molecular scale, the position of a reactive chemical, moving in a water-filled pore, depends on the tortuosity and connectivity of the pore-space, at a larger scale, and the accessibility of reactive sites to the molecule. Under unsaturated conditions, the ambient matric potential, which controls the spatial distribution of retained water and the thickness of water films within soil, presents an additional variable to consider, which may also impact on the reactivity and residence time of the molecule over a given time.

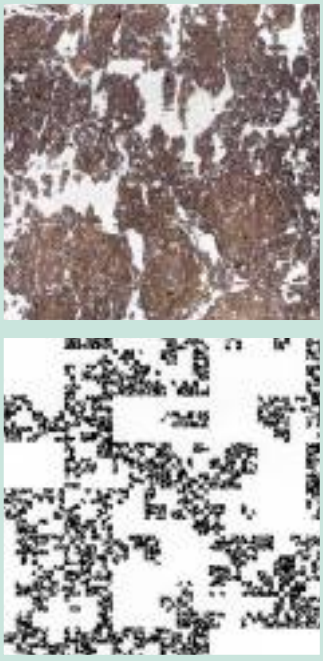

Figure 1 The link between complex soil architecture (top image) and fractal approximations (bottom image). Fractals capture the irregular and tortuous shapes that are ubiquitous in soil, and that impact on all soil processes.

Additionally, as we have shown in earlier work, the porosity of the soil can be scale-dependant; increasing as soil volume increases. The spatio-temporal heterogeneity of the architecture of soil thus impacts directly on the dispersal of pollutants and microbial contaminants to waterways and aquifers.

Accounting for complex architectures

The first step in our work was to describe mathematically the complex structures of all soils. The rough and tortuous shapes typically found are not usefully measured by classical geometry. From past work we have shown that fractal geometry can account for these complex structures, and has the added benefit of providing numerical approximations of the structural complexity that can be related directly to transport processes. Figure 1 shows how fractal geometry provides a useful description of real

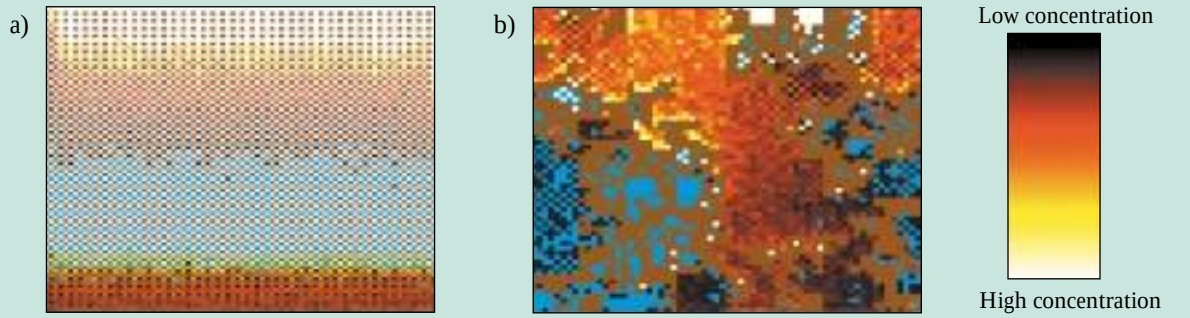

Figure 2 The effect of a heterogeneous physical structure on chemical adsorption. (a) Simulation of adsorption within a homogenous structure, and (b) adsorption within a heterogeneous fractal grid.

soils. By constructing model structures that account for complex pore shapes and connectivities, we can then use computers to simulate chemical, physical and biological processes. In this way we can test rigorously the hypothesis that the physical architecture of soil alone can have a significant impact on soil processes.

Linking chemistry and physics The ability of soil to supply nutrients to plants and retain potential chemical and microbial pollutants is crucial for food production and industries concerned with safe disposal of society's waste. The accepted protocols for measuring a soil's ability to adsorb and desorb chemicals (e.g. its cation exchange capacity) rely on destroying any natural physical architecture, through sieving and drying, and pouring chemicals onto the soil under saturated conditions. Therefore, the spatial relations between the chemical molecule, water, and the soil's natural architecture are completely lost. This leads to data based solely on artificial, structurally homogeneous, soil, bearing little relation to what really happens in the context of real soils.

When a solute is applied to such homogeneous structures, all reactive sites are accessible and adsorb solute until saturated (Fig. 2a). Essentially these methods give an adsorption capacity for the total reactive surface area of the soil. Clearly not all the reactive sites will be accessible to solute entering the soil. Site accessibility depends on both the soil structure and soil particle surface structure. We use a random recursive fractal lattice that accounts for the heterogeneity of the soil structure, and the random nature of soil particle surfaces (Fig. 1). When a solute is applied to this fractal lattice, a measure of the adsorption capacity of the accessible reactive surface of the soil is then determined. Figure 2b illustrates that the effect of structure on the chemical reactions is immediately apparent. It is interesting to note the occurrence of chemical 'hot spots' within the complex soil architecture, where solute has been trapped in, which are not seen in the simple architectures. This means that a proportion of solute is retained within the soil matrix rather than diffusing rapidly through the soil solution.

We use the Langmuir Adsorption Isotherm to express the proportion of occupied reactive sites at each solute

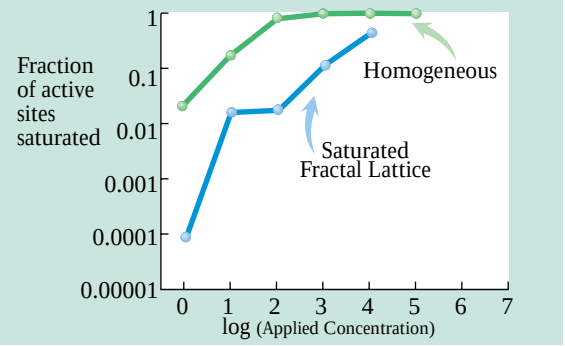

Figure 3 Langmuir adsorption isotherms for a homogeneous and structured 2D soil.

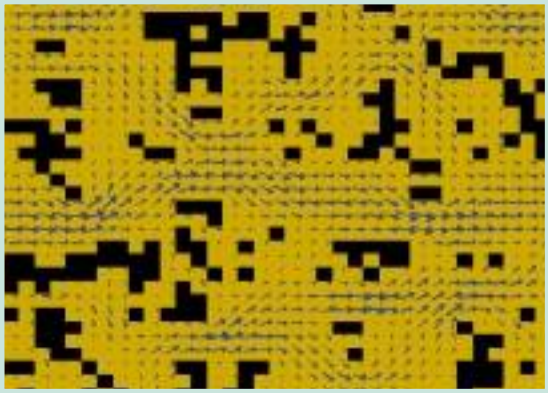

Figure 4 Lattice Boltzman simulation of solute flow through a random fractal lattice. The intensity and direction of flow are given by the blue arrows. Simulated soil is shown in black.

concentration, giving a measure of the adsorption capacity of the soil. Figure 3 compares the Langmuir adsorption isotherms for the unstructured, classical method and the structured, random recursive fractal lattice simulations. From these results it is clear that small-scale soil structure has a large effect on the adsorption of reactive solutes into soil.

Solute flow and structural complexity At the pore-scale, again using a fractal lattice as the basis of a complex architecture, we have modelled liquid and solute flow using the lattice Boltzmann equation discretized over a square lattice for 2D, and cubic lattice for 3D. Using the lattice Boltzmann algorithm, liquid flow is modelled by spreading a number of particles over the pore space within the lattice, which then move and collide according to the rules of conserving mass and momentum. We have shown that these simulations produce flow patterns entirely consistent with the solution of the classical Navier-Stokes equations that describe liquid flow at macroscopic scale. Two examples of simulated liquid flow for 2D and 3D soil structures using lattice Boltzmann algorithm are shown in Figure 4. From these simulations the major flow paths are clear. Additionally, areas where solute is present but little flow occurs are shown. These points show that only a relatively small portion of the pore space is involved in the main flow, and there are significant volumes where little or no flow occurs.

The two main areas of future research relate to the connection of the models for chemical reactivity and flow, which operate over two different scales, and to the development of the chemical model in 3D. In parallel we are conducting a joint experimental programme where we are developing methods which account for and measure the impact of complex soil structures on flow and chemical activity, with the ultimate aim of investigating the role of small-scale processes on flow and chemical activity at the catchment scale.

Whilst the examples illustrated in this article concentrate on physio-chemical processes, the concept of the rôle of the physical architecture of soil in controlling chemical processes, such as reactivity, is transferred easily to biological processes such as biocontrol, and pesticide efficacy.

Stable isotopes and biological processes

L.L. Handley, D. Robinson, C.M. Scrimgeour, R. Neilson & A.M. Johnston

In the last century, Kjeldahl's new chemical methods made it possible to measure the amounts of N fixed as NO_3^--N, NH_4^+-N and bulk organic N. Following World War II, the newly available enrichment techniques, using the stable isotopes of N, contributed greatly to knowledge about the movements of N in organisms and their environments. For some types of N studies, the use of highly ^{15}N-enriched tracers is still the preferred method. However, N cycling is complex, and many unresolved questions still exist. The only new way forward consists of using the natural abundance levels of $^{15}N/^{14}N$ ($\delta^{15}N$). At SCRI, we are pioneering new understandings arising from this novel tool while, simultaneously, investigating the mechanisms underlying the repeatable patterns of $\delta^{15}N$ found in plants, animals, soils and waters.

Work done at SCRI has established that types of soil N cycling, chiefly related to soil moisture availability and globally related to mean annual rainfall[1], explain a large proportion of the observed variation in the $\delta^{15}N$ of plants and soils. Field-based observations[2] suggest that, within sites, there may be a rank order of plant $\delta^{15}N$ which can be related to types of mycorrhizal association and, because of the role of these associations, to the chemical composition of the major soil N source. Even more recent field-based research[3] suggests that the within-site variation of plant $\delta^{15}N$ may be related to the interaction of mycorrhizal association with the availability of major plant nutrients such as N and P. Where nutrients are deficient, plants with ecto- and ericoid mycorrhizas appear to have much lower $\delta^{15}N$ values than those occurring in conditions of better supply. Controlled experiments with arbuscular mycorrhizal (AM) associations[4,5,6] have suggested that the species of AM-forming fungus interacts with external N concentration to increase plant $\delta^{15}N$. These data also suggest the possibility of using $\delta^{15}N$ to assess the effectiveness of mycorrhization in important commercial crops such as orchids and plantation forests.

The plants, alone, constitute another source of variation for plant $\delta^{15}N$. The extent to which plants fractionate the isotopes of source N upon assimilation of N and the extent to which plant N, having a non-average $\delta^{15}N$ value, is lost from the plant constitute other important lines of research within the Stable Isotopes Unit. In collaboration with the Cellular and Molecular Genetics Department, we have demonstrated, for the first time, an association between whole-plant $\delta^{15}N$ and plant growth. In a glasshouse environment, genotypes of wild barley (*Hordeum spontaneum* C. Koch) were exposed to stresses in the form of drought or nitrogen starvation. The potentially most productive genotypes (those which were most stress tolerant – in terms of growth and nitrogen content) had the most negative whole-plant $\delta^{15}N$ values. Under the conditions of this experiment, a more negative whole-plant $\delta^{15}N$ was equivalent to a greater discrimination against ^{15}N relative to the original source $\delta^{15}N$. Discrimination at a whole-plant level can occur only if some nitrogen is lost from the plant after being isotopically altered by metabolic processes. When measured in such experiments, whole-plant $\delta^{15}N$ may, therefore, reflect the extent to which nitrogen can be *retained* within tissues when plants are stressed, a feature of clear agronomic and ecological importance.

N transformations in soils affect plant $\delta^{15}N$ and generate N pools of interest to ecologists and environmentalists. In the Stable Isotopes Unit, we are using $\delta^{15}N$ of soil N and soil N pools to describe the spatial and temporal processes which generate NO_3^- in the soils and waters of the Ythan River Catchment, north of the city of Aberdeen. While the Ythan River is not

a source of potable water, it does exhibit seasonal blooms of the green alga, *Enteromorpha*. The interaction of river water chemistry and algal blooms is being investigated jointly with Professor John A. Raven (Dundee University).

It is conventional wisdom that excess amounts of NO_3^- (arbitrarily and by statute more than 50 mg/l) are responsible for nuisance algal blooms such as those found during the summer in the Ythan Estuary. This assumption is enshrined in EU legislation, requiring the designation of 'Nitrate-vulnerable zones'. Recently, this 50 mg-limit was extended to cover all surface waters, and an environmental monitoring network is being erected to measure the concentrations of NO_3^- in Scotland, England and Wales[7]. The new interpretations and techniques already being developed at SCRI constitute the only UK skill-base for complementing this monitoring with information on the sources of observed NO_3^- and for monitoring the extent to which the various, mandatory land management strategies ameliorate such NO_3^- generation.

Biotic stresses can cause plant $\delta^{15}N$ variations, which may become useful in understanding host-pathogen interactions. These interactions are inherently complex, and investigations are usually biased towards the effects of pathogens on plant hosts. Research within the Stable Isotopes Unit demonstrated that $\delta^{15}N$ could be used to detect physiological response(s) of a plant host to pathogen infection[8,9]. We followed-up this initial insight by investigating the effect of host on pathogen (using $\delta^{15}N$ and $\delta^{13}C$). Five species of plant-parasitic nematode from the family Longidoridae were transferred from their original host plants to seedlings of *Petunia hybrida* cv. Blue Picotee. After feeding on the new *Petunia* host for 28 days, three *Xiphinema* nematode species were ^{15}N-enriched and ^{13}C-depleted, compared with nematode populations that had fed solely on the original host plants. In contrast, no such changes were detected for the single species of *Longidorus* and *Paralongidorus* studied. Changes in whole body $\delta^{13}C$ are considered to be indicative of the new plant host (*P. hybrida*), whereas, differences in whole body $\delta^{15}N$ are probably related to the different feeding strategies used by the longidorid nematodes in this study. The techniques developed for this study could be applied to other soil microfauna, thus allowing investigation into the relationships that exist within the decomposer food web.

Underpinning all isotopic research at SCRI is state-of-the-art chemistry, new sample preparation techniques and development of new instrumentation.

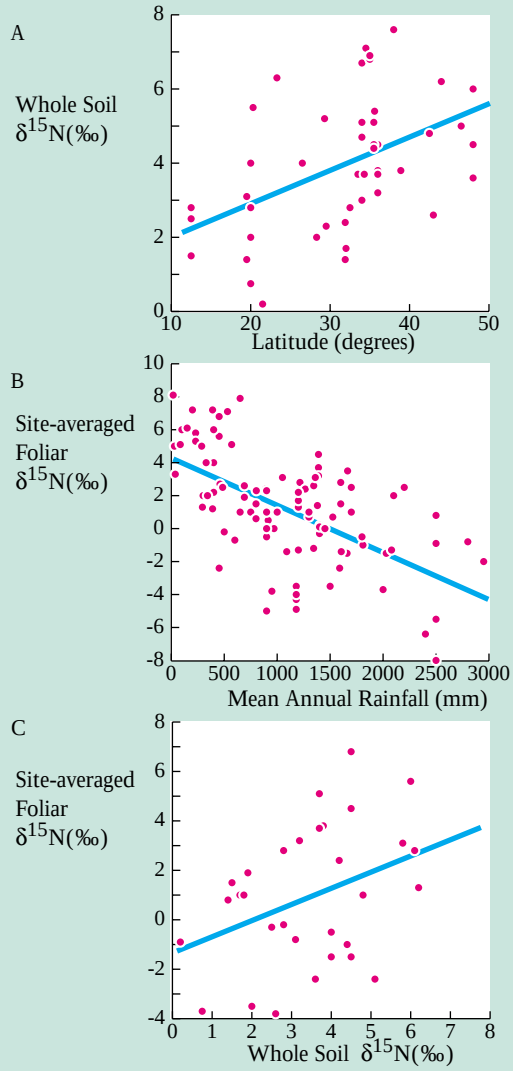

Figure 1 Globally, the $\delta^{15}N$ of soils and plants are correlated with mean annual rainfall or with latitude, for which rainfall is a major component. The relationship with site moisture reflects the variations of isotopically fractionating biological processes which are moisture-dependent.

The substantial increase in our understanding of natural abundance patterns of $\delta^{15}N$ [10] has been achieved by examining large data sets obtained by rapid, automated isotope analyses using state-of-the-art continuous-flow mass spectrometers. These instruments couple an elemental analyser to the isotope ratio mass spectrometer and operate under computer control. We have developed a number of robust analytical protocols for plant and soil samples using this instrumentation. Data from landscape-scale studies or from large sets of genotypes can be obtained readily and subjected to rigorous statistical analysis.

However, data from whole plant or soil samples do not tell the whole story, and increasingly we need information on particular chemical species within these samples. Separating individual compounds from a complex matrix such as soil is much more taxing than the simple drying, grinding and weighing of whole samples. Any separation must avoid loss or contamination which will alter the isotopic composition.

We are pursuing this problem for soil nitrogen species on two fronts. The first is the conversion to cleanly extractable organic derivatives, which can then be analysed using our high-throughput instruments. This approach is the most practical for studies requiring large data sets. The second approach is to use on-line separation, where components eluting from a gas chromatograph are analysed for isotopic composition. However, this approach will always be demanding of both operator and instrument time, and is best suited to detailed analysis of model systems which complement large field studies.

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Biomathematics and Statistics Scotland

Rob Kempton & Jim McNicol

Biomathematics and Statistics Scotland (BioSS) is devoted to the application of statistics and mathematics in the biological sciences. Its principal remit is to support the SOAEFD programme of biological research, which is mostly carried out within the SABRIs and SAC. This is achieved through a dispersed group of statisticians, mathematicians and computing experts based at BioSS centres in Edinburgh, Dundee, Aberdeen and Ayr. 1998 saw a continuation of our high output of publications and training courses. Success with competitive bids for new research funding allowed us to expand our research activity in two key areas: statistical genetics and bioinformatics; and veterinary epidemiology and food safety. Links were strengthened further with Scottish universities through new research grants and postgraduate students. The achievements of the BioSS unit at SCRI are highlighted below.

Work in statistical genetics received a major boost through BioSS staff winning two BBSRC grants in collaboration with SCRI scientists. One project funded under the initiative 'Genetics of Agriculturally Important Traits', is developing methods for linkage and QTL mapping in tetraploids using codominant markers. This will be used to enhance potato breeding techniques at SCRI. Work was completed on the development of statistical methods to detect mosaic sequences within multiple sequence alignments, which formed the basis of a SOAEFD studentship. BBSRC-funding, obtained in association with BITR and Department of Applied Computing, University of Dundee, will allow aspects of this work using hidden Markov models to be further developed and made more widely available. Our growing international reputation in statistical and computational genetics was acknowledged by invitations to present linkage analysis and QTL mapping workshops in Australia, and an EMBnet bioinformatics training course in Beijing.

Theoretical work on the use of additive models to analyse designed experiments in the presence of trends and local neighbour effects was completed and implemented in a set of Splus programs. Work on spatio-temporal models appropriate to semi-natural vegetation data is continuing in association with St Andrew's University and Institute of Terrestrial Ecology. The models have been applied successfully to Scottish vegetation land classification data.

Two projects have been undertaken with SCRI aimed at making current potato research more readily available to end users through decision-support systems. One project provides prediction of potato yields in the presence of cyst nematodes. The other involves a crop management tuber package for potatoes built around existing models of yield prediction and size distribution.

Analysis of codon usage patterns using graphical methods

F. Wright

Codon usage patterns The genetic code maps the 61 sense codons in protein-coding DNA to the choice of the 20 amino acids in the resultant protein sequence. Eighteen of the amino acids are coded for by more than one codon (Table 1). Three amino acids have six synonymous codons (e.g. Leucine with UUU, UUC, CUU, CUC, CUA, and CUG codons), five have four, one has three, and nine have two. Two amino acids (Methionine and Tryptophan) are each encoded by a single codon.

When molecular sequence data started to accumulate nearly 20 years ago, it was noted that DNA sequences that code for proteins did not randomly choose among codons that specify the same amino acid. Indeed the observed departure from uniform codon usage for each amino acid was a common feature. These observed patterns in synonymous codon usage were not simple and varied among genes within a genome, and among genomes. For *E. coli* and yeast genes, the main evolutionary force varying among genes was considered to be natural selection to optimise protein production (translational selection, acting on highly expressed genes), whereas with human genes, it was thought to be variation among chromosomal regions in the mutation process (biased mutation, resulting in base composition differences). It is only recently that sufficient DNA sequence data has accumulated to allow large scale studies of codon usage patterns in higher plants.

Studies of synonymous codon usage reveal information about the molecular evolution of individual genes and can provide data to train methods (genome-specific gene recognition algorithms) that detect protein coding regions in uncharacterised genomic DNA. Knowledge of codon usage patterns can also be utilised to design DNA primers and to detect horizontal transfer events.

Statistical analysis of genomic data and bioinformatics Earlier studies of synonymous codon usage were typically based on a sample of 100 genes from a genome. The ongoing eukaryotic genome projects are producing large volumes of sequence data and thus surveys of codon usage require to be automated. This will involve the automation of the extraction of protein coding DNA from the primary databases and of the subsequent statistical analysis of thousands of genes. In this article, some preliminary analysis of syn-

Phe	UUU	2	0.31	Ser	UCU	5	1.67	Tyr	UAU	0	0.00	Cys	UGU	0	0.00
	UUC	11	1.69		UCC	6	2.00		UAC	6	2.00		UGC	0	0.00
Leu	UUA	0	0.00		UCA	0	0.00	TER	UAA	1	3.00	TER	UGA	0	0.00
	UUG	23	2.56		UCG	1	0.33	TER	UAG	0	0.00	Trp	UGG	0	0.00
	CUU	13	1.44	Pro	CCU	5	1.67	His	CAU	1	0.33	Arg	CGU	14	3.50
	CUC	11	1.22		CCC	0	0.00		CAC	5	1.67		CGC	0	0.00
	CUA	2	0.22		CCA	1	0.33	Gln	CAA	5	0.28		CGA	0	0.00
	CUG	5	0.56		CCG	6	2.00		CAG	31	1.72		CGG	0	0.00
Ile	AUU	14	1.00	Thr	ACU	17	1.62	Asn	AAU	2	0.33	Ser	AGU	3	1.00
	AUC	28	2.00		ACC	18	1.71		AAC	10	1.67		AGC	3	1.00
	AUA	0	0.00		ACA	3	0.29	Lys	AAA	9	0.43	Arg	AGA	5	1.25
Met	AUG	6	1.00		ACG	4	0.38		AAG	33	1.57		AGG	5	1.25
Val	GUU	5	0.83	Ala	GCU	4	0.89	Asp	GAU	13	0.72	Gly	GGU	14	1.56
	GUC	7	1.17		GCC	13	2.89		GAC	23	1.28		GGC	5	0.56
	GUA	1	0.17		GCA	0	0.00	Glu	GAA	8	0.53		GGA	16	1.78
	GUG	11	1.83		GCG	1	0.22		GAG	22	1.47		GGG	1	0.11

Table 1 The codon usage table for an *A. thaliana* gene (Polyubiquitin; accession number L05361), consisting of 458 codons. Integer values denote usage of each codon; real numbers represent 'Relative Synonymous Codon Usage' (RSCU) values which are obtained by dividing the observed occurrence by the expected (e.g. the CUC Leucine codon occurs 1.22 times more often than the expected 9.0 assuming uniform usage of Leucine codons). The location of this gene on Figure 1 is at Nc=39.8 and GC3s=0.64.

		EXT-1		EXT-2	
Phe	UUU	0.53	(73)	1.26	(227)
	UUC*	1.47	(205)	0.74	(132)
Leu	UUA	0.27	(20)	0.88	(140)
	UUG	1.21	(90)	1.44	(229)
	CUU	1.55	(115)	1.63	(259)
	CUC*	2.39	(177)	0.65	(104)
	CUA	0.23	(17)	0.74	(118)
	CUG	0.35	(26)	0.65	(104)
Ile	AUU	0.88	(92)	1.28	(205)
	AUC*	2.00	(209)	0.69	(111)
	AUA	0.12	(12)	1.02	(164)
Met	AUG	1.00	(151)	1.00	(315)
Val	GUU	1.42	(153)	1.85	(320)
	GUC*	1.46	(158)	0.41	(71)
	GUA	0.29	(31)	0.70	(122)
	GUG	0.83	(90)	1.04	(180)
Tyr	UAU	0.36	(32)	1.32	(170)
	UAC*	1.64	(146)	0.68	(88)
TER	UAA	1.18	(11)	1.29	(12)
TER	UAG	0.32	(3)	0.43	(4)
His	CAU	0.65	(40)	1.52	(157)
	CAC*	1.35	(84)	0.48	(50)
Gln	CAA	0.69	(60)	1.03	(200)
	CAG*	1.31	(113)	0.97	(187)
Asn	AAU	0.36	(39)	1.23	(310)
	AAC*	1.64	(176)	0.77	(194)
Lys	AAA	0.59	(113)	0.92	(387)
	AAG*	1.41	(273)	1.08	(455)
Asp	GAU	0.93	(142)	1.51	(478)
	GAC*	1.07	(164)	0.49	(156)
Glu	GAA	0.71	(124)	1.02	(427)
	GAG*	1.29	(225)	0.98	(409)

		EXT-1		EXT-2	
Ser	UCU	1.58	(122)	1.87	(305)
	UCC*	1.41	(109)	0.39	(64)
	UCA	0.88	(68)	1.31	(214)
	UCG	0.53	(41)	0.50	(82)
Pro	CCU	1.35	(88)	1.85	(251)
	CCC*	0.89	(58)	0.33	(45)
	CCA	1.20	(78)	1.26	(171)
	CCG	0.57	(37)	0.57	(77)
Thr	ACU	1.30	(122)	1.58	(200)
	ACC*	1.56	(146)	0.52	(66)
	ACA	0.73	(68)	1.37	(173)
	ACG	0.41	(38)	0.52	(66)
Ala	GCU	1.75	(235)	1.89	(335)
	GCC*	1.42	(191)	0.47	(83)
	GCA	0.41	(55)	1.15	(203)
	GCG	0.42	(56)	0.49	(87)
Cys	UGU	0.90	(53)	1.25	(106)
	UGC*	1.10	(65)	0.75	(64)
TER	UGA	1.50	(14)	1.29	(12)
Trp	UGG	1.00	(55)	1.00	(126)
Arg	CGU*	2.21	(106)	0.64	(63)
	CGC*	0.88	(42)	0.12	(12)
	CGA	0.31	(15)	0.77	(76)
	CGG	0.17	(8)	0.58	(57)
Ser	AGU	0.47	(36)	1.09	(178)
	AGC	1.13	(87)	0.83	(135)
Arg	AGA	1.35	(65)	2.32	(228)
	AGG	1.08	(52)	1.57	(154)
Gly	GGU*	1.67	(333)	1.36	(245)
	GGC*	0.71	(141)	0.50	(89)
	GGA	1.44	(287)	1.38	(247)
	GGG	0.19	(38)	0.76	(137)

Table 2 Extremes (denoted EXT-1 and EXT-2) of the main trend from CA based on pooled codon usage from 28 genes (6273 and 10836 codons, respectively). Integers and real numbers are used as in Table 1. Preferred codons in the EXT-1 group are marked with an asterisk.

onymous codon usage among protein-coding genes in *Arabidopsis thaliana* will be discussed, using the 560 well-annotated genes analysed by Mathé[1] *et al.* This will serve as a pilot study of a higher plant genome, after which we intend to investigate the software requirements and statistical methods for the automated analysis of larger datasets.

Statistical methods can be split into two main types: (1) graphical methods that display the trends in the data, and (2) methods that explicitly test the significance of possible factors (e.g. base composition, protein expression level). The former approach is discussed here.

Graphical methods I: correspondence analysis An appropriate graphical multivariate method for count or proportion data is correspondence analysis (CA). Codon usage data from a sample of G genes can be arranged as a two-way contingency table, with G rows and 61 columns. CA can be used to extract the trends in this dataset, using either the raw counts (containing amino acid usage as well as synonymous codon usage information) or counts corrected for amino acid usage, i.e. relative synonymous codon usage (RSCU) values (see Table 1 for an example of RSCU values for a gene). Here we concentrate on trends among the genes. The CA of the RSCU values of the 560 *A. thaliana* genes produced a main trend that explained 11.2% of the variation. To illustrate this trend in synonymous codon usage, we have pooled codon usage tables of 28 genes that lie at either end of the trend (see Table 2). Initial inspection reveals that genes at

one end of the trend (EXT-1) tend to use synonymous codons ending in C, and that many of these genes are known to be expressed in large amounts. Similar CA results were obtained by Chiapello[2] *et al.*

The CA analysis produces other trends in the data. For brevity, we have only shown the main trend. Other multivariate methods can be applied to codon usage data. If distinct clusters are expected, then cluster analysis is an appropriate method. Cluster analysis was used by Mathé[1] *et al.* to partition the 560 genes in this study into two groups which, as might be expected, have similar codon usage to the patterns of EXT-1 and EXT-2, respectively.

Graphical methods II: the effective number of codons used in a gene Another approach to exploring patterns in synonymous codon usage among genes is to quantify, for each gene, the extent of the departure from uniform usage within each amino acid class. A commonly-used measure, the "effective number of codons used in a gene", N_c, was developed by Wright[3]. This produces a number, for each individual gene, that lies between 20 (when only one codon is used for each amino acid) and 61 (when all codons are uniformly used). N_c is based on the "effective number of alleles" (N_a) statistic from Population Genetics theory and is approximately the sum, over all amino acids, of N_a. This intuitive measure is essentially independent of gene length and a recent comparative simulation study[4] has shown it to be the best overall estimator of absolute synonymous codon usage bias. Nc can be plotted against factors (e.g. G+C content in the third codon position, GC3s) to investigate patterns of codon usage (this is analogous to plotting residuals against possible additional factors when carrying out a multiple regression analysis). Figure 1 shows the plot for the 560 *A. thaliana* genes in this study. The plot contains a reference line, labelled GC(ref), that shows the expected position of genes whose codon usage is only determined by variation in GC3s. This GC(ref) line is an approximate upper limit for the value of N_c. For example, if GC3s is zero, then only codons ending in A and T will be used, thus restricting the number of codons used to 30 out of the 61 sense codons. The position of genes lying well below the GC(ref) curve reveals that GC3s composition only explains some of the variation in *A. thaliana* codon usage.

Software for analysing synonymous codon usage patterns While correspondence analysis can be carried out on count data by most commercial statistics packages, CodonW (by John Peden; available from http://www.molbiol.ox.ac.uk/cu/Readme.html), a public domain program, will read in a set of protein-coding DNA sequences in FASTA format and carry out a range of analyses, including CA and the calculation of N_c and GC3s for each gene.

Production of codon usage data from DNA sequence data The preparation of error-free protein-coding DNA sequences as input to a survey of codon usage analysis can be time consuming. In particular, it is important to exclude coding regions that have been predicted by gene recognition algorithms, rather than by direct experiment. The Codon Usage Database[5] is a useful source of pre-calculated codon usage data, although it is not completely up-to-date with the primary nucleotide sequence database. Ideally, the permanent storage of codon usage data should be avoided and instead the codon usage table should be automatically calculated "on the fly" from each database entry. We are investigating the automatic extraction of such data from molecular sequence databases with the help of our colleagues at Bioinformatics and I.T. Research at SCRI.

Trends in *Arabidopsis thaliana* codon usage The amount of the synonymous codon usage variation

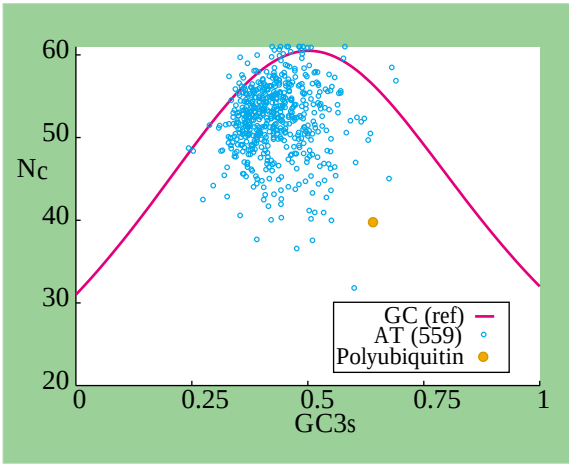

Figure 1 A plot for the 560 *A. thaliana* genes in the study.

explained by the main trend (11.2%) in this study of 560 *A. thaliana* genes is low compared to similar CA analyses[6] of human (32.5%), yeast (38.7%) and *E. coli* (30.5%). The main trend is partly due to variation in G+C (mainly C) composition among genes. The range of variation in base composition is not as extreme as found in Human genes, but is more extensive than found in yeast and *E. coli*. However, many of the high G+C genes are known to be highly expressed[2]. This confounding of G+C variation and protein expression levels complicates the interpretation of the analysis. Chiapello[2] *et al.* concluded that translation selection was the major factor, on the basis that intron and synonymous sites differed in G+C composition (if biased mutation was the main factor, we would expect that intron and synonymous sites would have similar base composition). Further analysis of this dataset may reveal other factors influencing codon usage. In particular, the chromosomal location of *A. thaliana* genes is a factor excluded from our analysis.

There are currently 6297 protein-coding regions in Nakamura's Codon Usage Database and more in the primary nucleotide sequence database. We intend to extend the analysis to this larger dataset once we have automated the analysis and improved the statistical analysis.

Future statistical work N_C is the best overall measure of absolute synonymous codon usage bias[4]. However, N_C did not perform well for very short genes (e.g. less than 200 codons long) where there are some amino acids that are unused, and we plan to produce an improved estimator of N_C.

Rather than use graphical methods to explore the data, we could use a Generalized Linear Model (GLM) to fit a model to the codon usage table. Such a model could be used to fit factors like G+C composition, chromosomal location, tissue-type, and gene expression level (if available). GLMs have not been applied to large scale surveys of codon usage, but have the potential to aid our understanding of the relative importance of factors. In particular, the interaction of mutation bias and translational selection in genomes with skewed base composition, e.g. *Dictyostelium discoideum* (mean genomic G+C approx. 25%) and *Streptomyces* spp. (mean genomic G+C 75%), is of particular interest.

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Research services

Administration Department

I.F. Harrington & I. Paxton

The Administration Department is responsible for not only the day-to-day smooth running of the Institute but also for longer-term assessments and planning. Its work includes two major areas, Personnel and Finance. Staff costs represent the major component of expenditure, accounting for 66% of the Institute's recurrent budget.

Not surprisingly, therefore, the work of the Personnel Section is vital to the success of the Institute. The Personnel Section carries out the administrative induction for new members of staff and supervises probation procedures during their first year of employment. It is responsible for monitoring absences with particular emphasis on those attributed to ill-health. Personnel handle requests for job evaluations at the request of both individuals and Institute management. In addition, it co-ordinates the recruitment of Institute Staff and, during the year, 46 posts were advertised, attracting 563 applications from external and internal sources. The training budget is administered by Personnel on behalf of the Institute Training Committee. Personnel staff collate training requests from staff and also arrange training with the appropriate training providers. Personnel are involved in monitoring the progress of the research students based at the Institute. During the year, approximately 50 students were working at the Institute. Assistance is provided to new staff, visitors and students with accommodation and other welfare related matters. Throughout the year, Personnel staff were involved with the Institute's Investors in People Initiative.

The Institute continues to operate under considerable financial pressure as the Grant-in-Aid it receives from Government has declined and will continue to decline in real terms. The control of the Institute's finances is therefore a critical activity to aid in the continuing production of successful science by the Institute. Capital grant received from SERAD is still an important source of finance.

However, increasingly, funds are gained from other sources to provide the necessary infrastructure to maintain and expand the quality and quantity of science within the Institute.

Each month the finance team within the Department pays in excess of 350 members of staff, processes in excess of 1100 purchase invoices from suppliers, and raises over 20 sales invoices. The Institute is project driven and over 200 projects are maintained and monitored at any one time. The team also maintains over 1500 items on its fixed asset register, ranging from PC Computers to laboratory buildings. The team is undergoing a period of rapid development as it introduces a more modern and flexible computerised accounting system, which will deliver a better service to the users of financial information.

Analytical facilities

W.W. Christie

Laboratory Accreditation

Within the Chemistry Department, the Gas Chromatography-Mass spectrometry Laboratories, Stable Isotopes Facility and Lipid Analysis Unit of MRS Ltd operate a formal Quality System, certified to BS EN ISO 9002 by SGS Yarsely International Certification Services Ltd. A generic Quality System operates in other parts of the Institute and this is summarised in the SCRI Quality Plan, a copy of which is included in the Institute's Corporate Plan. The measures required for implementation of the system are described in a Code of Practice document, a copy of which is issued to all members of staff. It is based on the correct maintenance of work records, in which specially designed hardback notebooks comprise the primary record, with other data recording systems, archival procedures *etc*. as secondary records. The preparation of written methods or protocols (Standard Operating Procedures) and the correct use of equipment and facilities are strongly encouraged. The plan ensures full compliance with all safety regulations, and demands high standards of laboratory hygiene. If required, the Quality System can be readily upgraded to the standard required for formal certification within any activity or area. The Chemistry Department operates an electronic archival facility based on the use of a compact disc (CD) writer installed in a personal computer. Data can be transferred over the network or from a portable high capacity data storage disc to the computer's hard disc, and then to CD. Each CD can hold up to 650 Mbytes of data, and two copies are made, one for the owner of the data and one for the archive.

Stable Isotope Facility

Stable isotopes are now basic tools for the study of plant physiology, crop genetics, ecology and food webs. Valuable information comes both from studying natural variation in stable isotope composition and from following the fate of added isotopic tracers. SCRI is equipped with a comprehensive range of modern instrumentation for stable isotope analysis. With these, we can tackle most of the biologically important low atomic number elements, ^{13}C, ^{15}N, ^{18}O and ^{34}S, in a wide range of solid, liquid and gas samples. All the instrumentation is based on continuous-flow isotope-ratio-mass spectrometers that are fully automated and operated through computer data systems. Automation allows a high through-put of samples, essential for many biological experiments where large data sets are required. For solid samples, the Europa Scientific Tracermass and 20-20 mass spectrometers are interfaced to Roboprep CN and ANCA-*NT* SL combustion sample converters. A Roboprep G+ gas purification unit is used for gas analysis. Plant samples of one to five milligrams are used, containing 25 to 100µg of the element of interest. Where possible, analytical protocols are devised to minimise sample preparation and fully exploit the automation.

SCRI also has expertise and resources for sample preparation from a wide range of matrices. These include plant sample drying and grinding, freeze drying and weighing facilities. Research support is aimed at developing new methods to assist the Institute's commissioned programme.

Mass Spectrometry

The Institute's three state-of-the-art mass spectrometers, which are devoted to structural analysis of organic compounds, continue to yield valuable information on a diverse range of materials pertinent to the research remit of the institute. The laboratory suite housing the instrument facilities will, in early 1999,

become part of a new integrated chemical facility which will include stable isotope mass spectrometry facilities, the MRS Lipid Analysis Unit and all of the Chemistry Department. The core instrument is a Hewlett Packard 5989B MS ENGINE research-grade quadrupole instrument with electron impact, chemical (positive/negative) ionisation modes and a mass range of 2000 amu. Distributed processing software permits off-line data processing and reduces analysis times. This instrument can provide mass and structural data on a wide range of organic compounds.

A further bench top instrument is dedicated principally to the analysis of naturally occurring volatile compounds. This consists of a Perkin Elmer automated thermal desorption system (ATD) linked to a VG TRIO-1000 quadrupole gas chromatograph-mass spectrometer and permits detailed characterization of the profiles of organic volatiles generated by biological systems. During the year, the gas chromatographic side was improved with the addition of a cold on-column injector. This should improve sensitivity of conventionally solvated samples and avoid undesirable degradation of thermally labile samples such as monoterpene alcohols.

A Finnigan SSQ 710C dedicated liquid chromatography-MS instrument, with atmospheric pressure chemical ionization (APCI) and electrospray ionization (ESI) interfaces, completes the facility. This has an ability to analyse samples whose high molecular weight, lack of volatility or polarity, precludes analysis on the other instruments. APCI and ESI are soft ionization techniques and generally only produce molecular ions, e.g. $[M-H]^+$ or MH^+, but the multicharge ionization mechanism of electrospray can extend the basic 2000 mass range of the instrument by a factor of about 20, giving a mass range of greater than 40,000 amu. This permits accurate mass determination of

peptides, proteins and nucleic acids to within 0.1%, compared to the 5.0% error usually expected from SDS-PAGE determination.

Mass spectrometric analysis at SCRI covers a broad spectrum of chemical investigations generated by the research programme of the Institute. A wide range of plant metabolites has been analysed, both in the native form and as derivatives, including sterols, monoterpenes, sesquiterpenes, pentacyclic triterpenes, dimeric forms of phenolic acids, glucosinolates, long-chain wax esters, peptides, essential oils, carbohydrates, polychlorinated biphenyls and lipids, including fatty acids. The facilities are operated by experienced and expert staff, ready to tackle and solve most structural problems.

The Institute's ISO 9002 certification now includes the analytical operation of both gas chromatography-mass spectrometry systems, with data archival facilities to appropriate levels, together with the required documentation, quality plan and standard operating procedures. Following a detailed survey and appropriate upgrading, we expect all of the important instrumentation to operate successfully at the start of the new Millennium.

Engineering and Maintenance Department

S. Petrie

The Engineering and Maintenance Department offers a technical design and maintenance service throughout the Institute. Preservation of Institute assets is of paramount importance and careful, skilled inspections are frequently carried out. Corrective maintenance work takes place to ensure the expected performance and life of equipment, vehicle, plant or building is achieved. The Department is divided into sections that specialise in a variety of engineering disciplines such as electrical, electronic, refrigeration, heating and mechanical engineering. It provides an engineering design and maintenance service to cover scientific and ancillary equipment, and building services, including heating, ventilation and air conditioning. There is also a farm workshop section providing maintenance facilities for a substantial fleet of tractors and agricultural machinery. The Department provides a general stores facility and a cleaning and security service. The workshops are generally well equipped to deal with the maintenance tasks assigned to them.

The rapidly changing and wide ranging scientific aims of the Institute ensure that laboratory alterations will always be a part of the Engineering Department's work. With this in mind, services to laboratories must be as flexible and adaptable as possible. Over the last few years, systems have been introduced which allow the Department to respond quickly and efficiently when changes are necessary, thus reducing laboratory disruption to a minimum. Scientists can now confidently bring new and diverse projects to the Institute knowing that a team is on hand to ensure the facilities will meet whatever requirement they may have.

During 1998, several areas of the Institute were refurbished to either enable new and expanded areas of work to be carried out or to simply improve the existing facilities. The main project undertaken this year centred around Building S.

Following a realignment of the various Scientific Units and Departments within the Institute certain Units found themselves located in different buildings to their new Departments. To overcome this, a plan of action was drawn up to solve not only this problem but also a general one throughout the Institute of insufficient laboratory and office space. The main element of the plan was to refurbish Building S, which was originally used for crops handling and storage.

During 1995, part of the building had been converted into a suite of laboratories for use by the Spectroscopy Section. Last year, the rest of the building was earmarked to accommodate the Institute's Chemistry Department and these areas have been or are now being converted into offices and laboratories. Part of the project involves the conversion of the loft area to form fully equipped and functional laboratories covering 215 m^2 of floor space to house the Institute's lipid research and the MRS Lipid Analysis Unit.

The majority of the work involved in this project has been undertaken by the staff within Engineering and Maintenance and includes the Electrical and Mechanical Services work, the telephone and IT network cabling, all joinery work including fitting out laboratories, and all painting works.

Once staff in the Chemistry Department are relocated, the space they vacate will be filled by various sections within the Cellular and Environmental Physiology Department and enable them to be housed together. This move in turn will free up sufficient space to allow all the various sections within the Soft Fruits & Perennial Crops Department to be accommodated within the same wing of another building.

Again, any changes or additions to the existing accommodation to allow the various sections to move will be undertaken by the Engineering and Maintenance staff.

The Department is also responsible for negotiating utility contracts with electricity, gas, water and telephone companies and successful negotiations in previous years have now made further savings in these areas

difficult. The electricity unit cost was held at around the same level as last year's but market forces resulted in our gas costs rising. Telephone call charges were reduced but water charges have escalated due to a combination of increased unit costs and various forms of relief being withdrawn. Costs in this area are due to increase far beyond the rate of inflation and although water usage will again be addressed, costs will inevitably rise.

The Department monitors usage and efficiency of all the four utilities and although there will always be room for improvement, levels of use are now unlikely to fall significantly without major capital investment.

In recent years, ever increasing and more demanding legislation has had an effect on the work and the working practices of the Department. The Institute must, and does, provide a safe working environment for its employees and visitors and, at the same time, endeavour to stay clear of litigation, but the cost of doing so is increasing annually. Much of the work to ensure their safety goes unseen by the majority of staff and often there are no tangible benefits to be gained from it. With the severe financial difficulties being faced by the Institute, it would be easy to become complacent in this area and it is to the Institute's credit that it continues to find the necessary resources to fund this properly.

We are also educating scientific staff into understanding that legislation now clearly defines areas of work, such as those associated with electrical and gas systems and equipment, that they cannot enter into without undermining the Institute's legal position. It is sometimes difficult for non-technical staff to understand that simple tasks such as changing a 13 amp plug or fuse must always be carried out by the relevant trained person within the organisation.

More and more time, effort and resources are being spent to cope with the 'what if' scenario, all of which add to the ever increasing workload of the Engineering and Maintenance Department.

Estate, Glasshouse & Field Research Department

G. Wood

A new SCRI committee – the Genetically Modified Crop Field Release Committee – has been established to oversee all GM crop field releases at this Institute. The committee will ensure compliance with the requisite rules and regulations and that the associated new operational practices are in line with the SCRI standards. It extends the control over GM research exercised by the Institute's Genetic Manipulation Safety Committee. The committee ratified a straightforward procedure that must be adhered to when SCRI is involved in any work which includes the deliberate release of a genetically modified crop. This procedure is over and above the statutory rules, regulations and procedures governing the deliberate release of GMOs, laid down by the Department of the Environment Transport & the Regions and the Advisory Committee on Releases to the Environment. The new committee met four times in 1998, to consider matters which included the monitoring of previous release sites and a new application (98/R23/5) for consent to release GM potatoes.

During 1998, there were also several meetings involving representatives of the local Community Council, Perth and Kinross Council, and SCRI. The successful outcome from this constructive co-operation was the recognition of official rights of way and additional permissive routes for pedestrians through the Mylnefield site. It is hoped that the routes will be 'named' following a competition sponsored by the Community Council and open to children attending Invergowrie Primary School.

Under the supervision of David Petrie, members of staff in the Field Research Unit, in conjunction with L Dixon Associates, undertook the installation of a major irrigation/fertigation system in nearly 4 ha of newly planted raspberry trials. Underground feeder and lateral pipes were connected to the existing underground ring-main fed with water from the bore hole. Pressure reduction and dilution feeder (with bypass) components were incorporated. Drip-lines were attached to the bottom wire of each row of raspberries. The system will be used to aid the quick establishment of the plant material during that critical phase immediately post-planting and during the subsequent cropping and selection seasons.

Further investigations were undertaken by Peter Gill and Barry Robertson in the Glasshouse Unit concerning the auto-control of the environment and the usage of space within glasshouses. With a physically limited facility such as glasshouses, facing steadily increasing pressures from various research programmes to cope with more and more plant material, it is vital that the efficient utilisation of such a resource is maximised. The data collected over a full season showed that there were some areas of the glasshouse facility where the effective usage of space might be improved and inputs reduced. A TOMTECH HC160 auto-environment control computer, linked to external (meteorological station) and internal sensors, delivered a highly integrated control of lighting, heating, venting, and internal thermal/shade screens in a wing of Cambridge glasshouses. This system also delivered overall reductions in energy usage and hence savings in running costs in the order of 30% (confirming previous data; Ann. Rep. 1996/97, 203.). These take no account of the additional savings in man-hours compared with manually operated, non-inte-

grated systems. As important, was the fact that the environment for the plants was more stable, and hence the quality and uniformity of the material were improved. The TOMTECH system, which is less complex than some other systems, proved entirely capable, reliable, very straightforward to operate, and delivered the degree of environment control required. We have a great deal of catching up to do since we need such a system installing in several other areas of our existing glasshouse facility. The initial one-off capital outlay is more than balanced by the immediate and continuing savings in running costs, the better quality plants and hence the better quality science!

This Department's commitment to staff development and training (Ann. Rep. 1995) continued through 1998. One of the practical benefits gained from the implementation of a staff training and development programme, in line with the Investors in People initiative, was that colleagues who had already undergone the appropriate multi-skilling training were able to cover some of the additional duties and areas of work in the immediate short-term following staff changes. In the spring of 1998 three new appointees were inducted into and immediately included in the ongoing training and development programme. The service/support rôle of this Department is crucial to the success of the Institute's science programmes, and it is vital that we attempt to maintain and provide for a continuity of skill level and experience. With financial support from Scottish Enterprise Tayside, together with inputs from Agenda Training Ltd and the Carse of Gowrie Training Group, we took on a young Skillseeker under trainee status for a Scottish Vocational Qualification in Extensive (field) Crop Production. Mark Orchiston progressed well and after 10 months he transferred to a Modern Apprenticeship (SCRI's first) under employed status. SVQs assess the skills and knowledge gained through work and training. A full SVQ can take between 9 months and 3 years to complete and each covers a complete work rôle. It is made up of a number of units, some mandatory and some optional. The majority of the training and assessment is delivered in the workplace. The system means the awards closely reflect the individual's work and the level of competency required to do it. The level of competency is measured against nationally recognised, quality assured standards. Again with financial assistance from SET, four senior members of the Department's staff successfully undertook and were awarded the D32 Assessor qualification (a Training and Development Lead Body award). This equips them to undertake competency and skills assessment within the SVQ system. This will be put to direct use in the next phase of our training and development programme with some members of the existing staff already undertaking SVQs.

Scientific Liaison & Information Services

W.H. Macfarlane Smith

The Department of Scientific Liaison and Information Services (SLIS) continued to publicise and promote the science of SCRI whenever cost-effective opportunities arose. Events such as the British Potato Council's 'Potato Technology '98' provided an ideal vehicle for doing so. The Institute's national and international reputation rose ever higher and had a direct consequence in the increasing numbers who visited SCRI. International visitors formed an increasing percentage of the total, with parties from distant countries such as Argentina, Estonia, New Zealand and Palestine among the many who came to SCRI in the past year.

Visitors from the International Congress of Plant Protection on the post-congress tour of the Institute.

Universities continued to take a great interest in the Institute's work, with staff and students from the Universities of Aberdeen, Dundee, Edinburgh, Glasgow, Heriot Watt, Newcastle, St Andrews and Strathclyde among those who visited regularly. It was a particular pleasure to welcome the new Secretary of The Scottish Office Agriculture, Environment and Fisheries Department, Mr John Graham, soon after his appointment, to demonstrate to him a range of the Institute's research activities. An increased effort, in collaboration with MRS, has been put into bringing to SCRI the individuals and organisations who wish to know more about the Institute's work and, thereafter, may wish to invest in our R & D. Links with the less-advantaged countries have not been neglected, with visits by individuals and groups from countries such as Bangladesh, China and Greece.

Our involvement with local organisations such as Bio-Dundee helped to create a critical mass to promote the biotechnological R & D undertaken by groups in and around Dundee. SLIS was also a founder member of the new ECRR Public Relations Group. New developments such as the Dundee Science Centre and the Carse of Gowrie Initiative (to revitalise industry, tourism and transport links) provided opportunities to inform the public about SCRI's work and be informed about areas of mutual scientific interest. The need to convey easily understood information about the Institute's research to the general public is an increasing precondition of Government and European Union support. To this end, staff have given interviews to the press, radio and television, and made presentations to a wide range of clubs and societies, from Citizens Associations through Business Clubs to Rotary and Probus.

Lord Provost of Dundee, Mervyn J Rolfe, J.P., with Dr Asitava Basu and Mr Maloy Ghosh, visiting students from Bangladesh, at the City Chambers.

The Unit of Bioinformatics and Information Technology , in collaboration with the Engineering and Maintenance Department, has continued the programme of upgrading and extending the computing facilities at SCRI. The Institute is committed to phasing out all computers with a 386 (or less) processor by the year 2000

and over- seeing a wholesale switch to Windows NT and Office 97 software in the same period. A major effort, involving staff from the BITR, Library and Visual Aids Units, has been put into upgrading the Institute's web pages (http://www.scri.sari.ac.uk) and creating an Intranet as an easily accessible source of 'in house' information, with pages dedicated to Staff, Administration, Health and Safety, Engineering, Library, Appointments, etc.

The Library continues to be under pressure, with costs escalating well in excess of inflation. Again, the donations of books and other material made by members of staff are gratefully acknowledged, as these allowed the Library to make optimum use of its funding. Nevertheless, difficult decisions had to be taken again about the cancellation of some journals. The rapidly growing collection of links to electronic journals and the catalogue of useful and interesting web sites, reached from the Library Intranet page, have increased rapidly in popularity. The process of re-locating little-used stock has continued and plans have been drawn up for the Library's further upgrading, including additional shelving and computer terminals. Currently, discussions are taking place about the difficulties which the Library would face if existing links with the University of Edinburgh have to be severed. This would increase annual running costs by at least £10K.

The Visual Aids Unit continues to provide a wide range of services in photography and graphics. The unit's output increased yet again, as a result of the increasing number of staff, students and visitors who required routine services. In addition, the greater demands to publicise the Institute's science and commercialise the products of its research, have resulted in increasingly elaborate PR material and displays, the production of videos, etc. A measure of the high standards of the unit's work was shown by awards of a Silver Gilt Medal for the Ladybird Spot Checks exhibition at the Dundee Flower Show, Best Poster at the ICPP conference in Edinburgh, which was attended by 2000 delegates, and 1st and 2nd prizes at the Scottish Microscopy conference. On a commercial note, 300 copies of the in-house 'Ladybird Video' have been sold.

The Health and Safety group has again had a vital rôle to play in the successful undertaking of complex and potentially hazardous research procedures. All the research and research support activities upon which SCRI is engaged have been subjected to health and safety risk assessments. The procedures and substances used in these activities are extremely diverse and risk assessment places a significant management burden on scientists and support staff. Meetings of safety co-ordinators in the BBSRC and SABRI institutes and SAC took place throughout the year with SCRI represented at the joint BBSRC/SABRI meeting at BBSRC Headquarters, Swindon, the SABRI meeting at MLURI, Aberdeen and the ECRR meeting at SAC, Edinburgh. These meetings are able to draw upon the support of safety personnel in BBSRC's Human and Corporate Resources Group. This unit circulates Hazard Notices, referring to work activities and substances, Health and Safety Information Notes, which highlight new legislation and general points of interest, and Policy Notes, which provide guidance in Health and Safety issues for institute senior management teams. A comprehensive pack, giving guidance on the risk assessment of substances, has recently been received from BBSRC and the procedures are being integrated into SCRI's system of health and safety management. The implementation of such policies will improve progress toward standardisation in health and safety management across the biological PSREs, which will in turn simplify the processes involved.

Reception, as the first point of contact for either visitor or telephone caller, continues to have a vital rôle in public perception of the Institute. It was a pleasure to welcome Louise Fiddes to the staff and to see her maintain the high standards of efficiency and warmth of welcome for which the Institute is noted.

All staff in SLIS, in their different rôles, played a major part in preparations for and the presence of the Visiting Group, especially as the first series of dates identified for the visit were postponed. Due to staff changes, etc., this resulted in the preparation of virtually a complete new set of documents. It is a measure of SLIS skills and commitment that this was achieved so quickly and efficiently.

Media Kitchen

W. Ridley

The Media Kitchen was established in 1996 to provide a wide range of sterile microbiological, mycological, plant tissue culture, media preparation and disposable plasticware for the Institute's laboratory staff.

The Media Kitchen operates as a research facility under the central administrative overhead, to minimise bureaucracy; nevertheless, each user site is 'shadow-tolled' for its throughput of consumables,

etc.

Buying 1.5 million Eppendorf tips and 0.5 million Eppendorf tubes per year in bulk has resulted in savings of over 50%. The media kitchen frees the innovative scientists, visiting workers, trainee students and support technicians from the repetitive and time-consuming tasks associated with the above.

The Media Kitchen is staffed by two full-time and two part-time workers and the facility is supported and enhanced by the efforts of Walter Burry and James McMillan who were recruited through the Helm Project in Dundee.

The support staff fill, on average, 275 tip boxes and 50 Eppendorf tubs per week and deliver media to 12 pick-up and drop-off locations around the site on a daily basis. They also collect and recycle the glassware and collect, autoclave and dispose of waste microbiological materials.

Changes in the Media Kitchen annual turnover are shown in Figure 1.

	1997	1998	
Tips (boxes of 100)	13,933	14,300	+ 3%
Eppendorf tubs (c.200 tubes/tub)	2,600	2,620	+ 1%
Agar plates	37,011	43,600	+18%
Other items (bottled & capped)	24,654	45,080	+83%

Figure 1 Media kitchen outputs 1997 and 1998.

The success of the Media Kitchen indicates that a larger working area and an increase in staff will be necessary to meet the ever-increasing demand for this essential core facility. Given the large number of visiting scientists and students (of all standards and backgrounds) who work at SCRI, the provision of a standardised, quality assured media and sterile disposable-ware facility, with its daily delivery service and daily removal of waste microbiological materials, has proved invaluable both to researchers and those monitoring costs and assessing value for money.

Mylnefield Research Services Ltd

N.W. Kerby & J.B. Snape

Mylnefield Research Services Ltd (MRS) was established in 1989 as the commercial arm of the Scottish Crop Research Institute (SCRI) to enhance competitiveness, and to understand and fulfil the needs of industry. MRS not only markets the resources and expertise of SCRI, but also undertakes near-market research and development. MRS places particular emphasis on developing partnerships and strategic alliances with industry.

MRS acts as the gateway to a variety of skills unique within the UK biological, agricultural and horticultural research services, ranging from fundamental studies on genetics, molecular biology and physiology, through agronomy and pathology, to glasshouse and field trials from a single site. As a technology transfer company, MRS is able to market the scientific expertise and resources of SCRI, and promotes the contribution of science and technology to wealth creation and the quality of life.

Mission Statement

Mylnefield Research Services Ltd will exploit commercially the scientific expertise and resources of the Scottish Crop Research Institute while protecting its charitable status and intellectual property.

The aims of MRS are entirely consistent with the Government's policy objective of improving the contribution of publicly-funded science to wealth creation and ensuring the productive use of Government assets.

The commercialisation activities of MRS take a variety of forms and include: research and development collaborations with industry; licencing of technology to third parties; provision of services and consultancy. A future objective is the creation of joint ventures and spin-out companies which is consonant with the Scottish Executive Strategy for Agricultural, Biological and Related Research 1999-2003.

The calendar year 1998 was characterised by major mergers of seed and agbiotech companies and this has continued into 1999. The driving force for these mergers and alliances between agrochemical multinationals and specialist genomic companies is the acquisition of intellectual property (IP), freedom to operate, and realising a clear route to market.

Roles and Responsibilities of MRS

MRS provides a service to SCRI scientists by

- Providing information on funding opportunities
- Assisting with the preparation of grant applications and research proposals
- Costing and financial management of programmes
- Negotiating contracts
- Project management
- Identifying and protecting IP
- Searching for 3rd Party IP
- Managing IP
- Writing business and implementation plans
- Marketing SCRI's scientific expertise
- Developing new markets for SCRI's and MRS's IP
- Licensing
- Diversifying the funding base
- Promoting SCRI as a centre of scientific excellence

Finances

From the time of incorporation, MRS has been self-sufficient in providing its own accommodation and staffing, achieved without start-up funding, Government subsidy or venture capital.

The turnover in the financial year 1997/1998 was £1.79M, of which almost £1.16M was used to directly fund research projects at SCRI. Income was generated through royalties (6%, previously 3%), analytical services and consultancy (5%, no change), contract research (45%, previously 47%) and collaborative research (44%, previously 45%).

Personnel

During 1998, Dr Julie Squires was appointed to work on a commercially-funded project related to soft fruit improvement and disease control.

Intellectual Property

MRS has continued to manage SCRI's intellectual property portfolio. Three new preliminary patents were filed in the financial year 1998/1999. The first covers a method for the identification of strains of *E.coli* O157 (Toth and Hyman) which evolved out of work on *Erwinia*. This methodology is currently

being validated by the Scottish Reference Laboratory for *E. coli* testing in Aberdeen. Tissue-specific promoters (Machray, Hedley, Davidson) were also the subject of a patent application and have attracted interest from several commercial companies. Finally, SCRI's patent portfolio in the field of viral vectors was strengthened by the filing of a patent that covers the applications of a transporter protein (Talianski, Riabov, Robinson and Wilson) derived from umbravirus. This complements the Overcoat® virus vector patent (Chapman, Santa Cruz, Oparka and Wilson) which was granted in Australia and New Zealand in 1998 and is pending in the USA, EU and Canada. Another patent granted in 1998/1999 was the spliceosomal promoter patent which was granted in Australia, New Zealand and the USA.

Training

Dr Jonathan Snape, Commercial Manager of MRS, was awarded a Sainsbury Management Fellowship in the Life Sciences by the Gatsby Foundation. This award, the first in Scotland, covers the fees for studying a MBA by distance learning at the University of Strathclyde, and will strengthen the management team at MRS.

Marketing

MRS has continued to market vigorously the expertise and facilities available at SCRI. In addition to a number of new brochures and flyers highlighting specific areas of expertise, MRS has launched a company website which has attracted a great deal of interest and potential customers primarily from North America. MRS has been represented at BIO 98 in USA, ABIC 98 in Canada and numerous other conferences and exhibitions in the UK. The number of commercial companies visiting SCRI in 1998/1999 increased significantly, leading to several possible collaborations.

Royalties

Royalty income increased by more than 30%, primarily due to the success of the strawberry cultivar Symphony which performed exceptionally well, both in the UK and The Netherlands. The blackberry cultivar Loch Ness continued to perform well, as did the raspberry Glen Ample. Potato royalties were disappointing, and several cultivars have been dropped by our commercial partners. However, royalties from brassicas (marketed by Advanta and Nickerson), in particular Caledonian kale and the swede varieties Invitation, Airlie and Kenmore, were higher than expected.

Lipid Analysis Unit

The MRS Lipid Analysis Unit continued its steady growth and expanded its portfolio of services. A major move into the analysis of serum fatty acids has opened up a potentially very large market, and has necessitated the recruitment of another technician in 1999. The core business remains the analysis of evening primrose and borage oils.

Collaborative Research

MRS has continued to be involved in the project management of four Link Schemes and two EU projects, and has assisted SCRI scientists in the preparation of proposals for funding from the EU and other sources. In 1998, eight EU contracts, with a total value of almost £950k were secured. The levy boards continued to support the research at SCRI; contracts with the BPC worth £241k (including one project jointly funded with SOAEFD) and with HGCA worth £31k were signed in 1998.

Contract Research

Nine new contracts were signed with commercial partners in 1998, including a 3-year, multi-million dollar contract with Biosource Technologies of California, USA, to develop viral-vector technologies and eight other contracts worth a total of almost £1M. These contracts cover a wide range of topics including lipid analysis, plant breeding, plant biochemistry and genomics, and include several well-known multi-national companies that can not be named for reasons of confidentiality.

Acknowledgements

MRS gratefully acknowledges its sponsors and the co-operation of SCRI scientists and administrative staff for their contribution to the success of the company.

Scottish Society for Crop Research

D. L. Hood

Trustees:	Mr A G M Forbes Mr G B R Gray Mr I E Ivory Mr A Pattullo
Chairman:	Dr D A S Cranstoun
Vice Chairman:	Mr J M Drysdale
Members of Committee of Management:	Mr J Arbuckle Mr A C Bain Mr D Craib Mr A Logan Mr G Rennie
Secretary and Treasurer:	Mr D L Hood

Registered Office: c/o Scottish Crop Research Institute, Invergowrie, Dundee DD2 5DA

Membership Numbers: 300

The Scottish Society for Crop Research is a registered Friendly Society, formed in 1981 by the amalgamation of the Scottish Society for Research in Plant Breeding and the Scottish Horticultural Research Association.

The Society provides a link between the Scottish Crop Research Institute and farmers, processors and other interested bodies:

* by organising field walks and meetings for the exchange of information;
* by financing science based publications for the benefit of the membership;
* through the formation of crop based sub committees which maintain contact with members on specialised topics.

In recent years the nature of public funding has changed, so that it now primarily focuses on scientific research that is remote from the farmer and consumer. As a consequence, the Society is directing some of its funds for work to be undertaken which will have a more direct relevance to the farmer.

The Crop Sub Committees of the Society have presented proposals for various works of this nature and approval has been given for one project to commence. Entitled "Malting Quality Analysis in Winter Barley Mixtures", this research is set to generate interest among various sectors of the industry.

D.L. Hood (l), S. Santa Cruz, J.W. Crawford, D.A.S. Cranstoun.

The Peter Massalski Prize, a biennial award for the person(s) under 36 years old who is/are considered to have done the most meritorious research while working at SCRI, was jointly awarded in 1998 to Dr John W Crawford and Dr Simon Santa Cruz. The Society provides administrative support for this Prize, which was established in memory of Dr Peter R Massalski by his parents Professor and Mrs T B Massalski.

Members of the Society and friends will be sorry to read that Mrs Massalski died on the 19th February 1999 after a long illness. As a result of the continuing generosity of Professor Massalski and an additional anonymous donation towards the Massalski funds, the Society Trustees have increased the shareholding whose dividends provide the monies for the Prize.

The Society Soft Fruit Walk continues to provide a focus for the industry. Likewise the Potato Crop Open Day, in conjunction with the British Potato Council,

the Scottish Agricultural College, and the Institute, has seen increased numbers attending and commenting favourably on what has been exhibited. Suggestions for improvements have been incorporated in the forthcoming 1999 presentations.

The Soft Fruit Sub Committee was particularly active during the year, and there were also meetings organised by the Potato and Cereal Sub-Committees.

The Management Committee met twice during this year, in May and November, and were pleased to have the guidance of the Trustees for these meetings. They were concerned to note the ill health of the Director, Professor Hillman, and the Chairman of the Society at the Annual General Meeting in 1999 extended the best wishes of the Society for a speedy recovery to Professor Hillman.

Society membership numbers have declined over the decade in step with the decline in the farming community. Challenges facing the industry as the new Millennium dawns are considerable, yet Society members are to the forefront in new technology and its applications and welcome the opportunity to present their findings to their fellow members when the occasion arises.

The Society Annual General Meeting has, in the past, provided an opportunity for distinguished academic, media, and governmental representatives to express their views on topics of interest to the agricultural and horticultural membership of the Society.

The Management Committee welcomes suggestions for research topics and comment from members and others, and urges the latter to contribute to the Society by becoming a member, and thereafter possibly joining one of the Crop Sub Committees or indeed the Management Committee.

A Newsletter is distributed to Members and media, and contributed articles, together with photographs of interest, should be forwarded for the attention of the Secretary and Treasurer.

Staff Association

D. Guy

The Scottish Crop Research Institute has a very active Staff Association, with approximately 230 members. The primary aim of the Association is to raise money for a chosen charity, and this is achieved through raffles, functions, prize draws and donations from companies.

The chosen charity for 1998 was The Mackinnon Centre, a purpose-built 12 bed respite unit, set in its own grounds at Broughty Ferry, to the east of Dundee. The Centre's Physical Disability Resource team provide a service as part of the Social Work Department's community care provision in Dundee. They help local people, aged between 16 and 65, who have a physical disability or a progressive illness. A cheque for £909 was presented to Mr Dave Mackenzie and Mrs Mary Anderson from the Centre.

Staff Association members also support other causes. SCRI scientists scaled new heights in their efforts to help provide water supplies for people in some of the world's poorest communities. Alastair Stewart, Chairman of the "WaterAid Munro+ Challenge '98" Steering Committee visited SCRI to collect a cheque from the 17 scientists who between them reached the top of six Munros lying between Blair Athol and Skye - namely Beinn a 'Ghlo, Sgurr nan Eag, Sgurr Dubh Mor, Beinn Bhrotain, An Sgarsoch, Sgurr Alasdair - and who raised nearly £800 in sponsorship in the process.

The Staff Association organises a number of outings, including: golf competitions; go-karting; windsurfing; gorge walking, and hillwalking. It also runs a weekly aerobics class, and provides the strips for the Institute's football and netball teams.

David Guy, Chairman of the Staff Association (r) and Jane Fairlie (l), Secretary, hand over a cheque to Dave Mackenzie and Mrs Mary Anderson from The Mackinnon Centre, the chosen charity in 1998.

David Guy and Rhonda Meyer present a cheque to Alastair Stewart, with members of SCRI staff.

For the families of Staff, it hosts an annual Summer BBQ, a Christmas Ceilidh and Disco, and an annual Childrens' Christmas Party.

There is a monthly draw for cinema tickets and a meal for two, as well as periodic Prize Draws. Two popular prize draws during the year included tickets to Murrayfield, generously donated by the Scottish Rugby Union, and Les Miserables.

The Staff Association organises a number of discounted deals for its members, including Corporate Sports membership at a reduced price at Dundee University Sports Union, and purchase of seeds and plants. The Association also holds Corporate Membership of the National Trust for Scotland, which provides five membership cards that are available for use by Staff, and provides Which Magazine for the SCRI Library.

Membership of the Staff Association is £1.50 per month, and an AGM is held annually, at which time office bearers and the committee are elected, and the charity of the year is chosen. The charity chosen for 1999 is Cystic Fibrosis.

Publications

Publications are classified in the following manner:

J	Papers describing original research in refereed journals.
R	Critical reviews in journals, book chapters and reviews in books - providing each has been edited externally.
P	Published proceedings of contributions to conferences or learned societies (including published abstracts).
T	Technical reports, other publications.
O	Popular articles, other publications.

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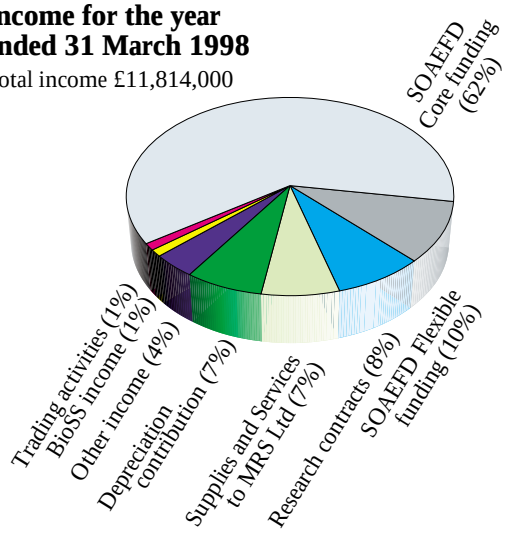
Summary of the Accounts

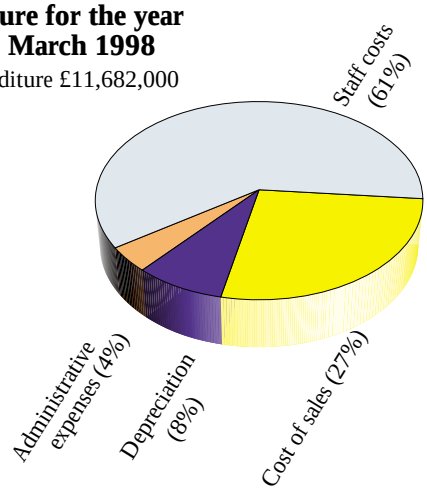
Income for the year ended 31 March 1998

Total income £11,814,000



Expenditure for the year ended 31 March 1998

Total expenditure £11,682,000

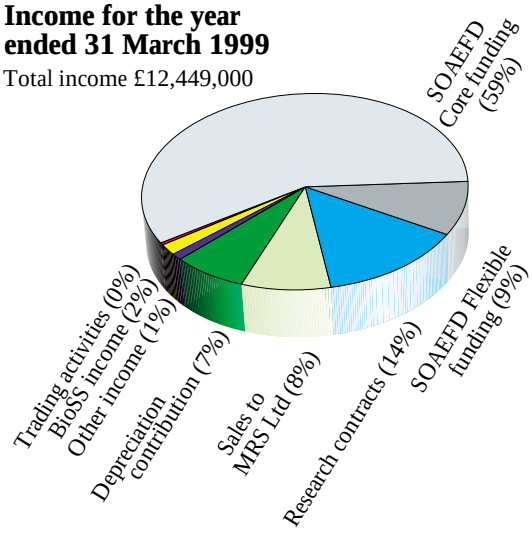


Balance sheet at 31 March 1998 **Total value £12,457,000**

Assets		Liabilities	
Fixed assets	92 %	Capital reserve	90 %
Stocks	1 %	Income & expenditure account	3 %
Debtors	7 %	Current liabilities	7 %

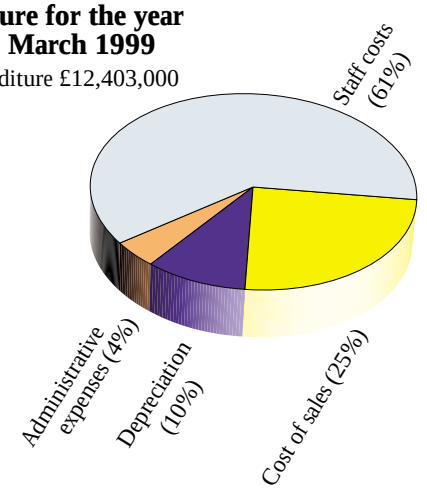
Income for the year ended 31 March 1999

Total income £12,449,000



Expenditure for the year ended 31 March 1999

Total expenditure £12,403,000



Balance sheet at 31 March 1999 **Total value £19,247,000***

Assets		Liabilities	
Fixed assets	94 %	Capital reserve	93 %
Stocks	0 %	Income & expenditure account	3 %
Debtors	6 %	Current liabilities	4 %

* following revaluation of the SCRI land and buildings

The Governing Body

Left to right, Back row : B. King, R.J. Cogdell, Sir John S. Marsh, P. Whitworth
Front row : K. Hopkins, J.E. Godfrey, J. M. Drysdale
(Absent: J.J.F. Belch, J. Evans, J.M. Sime, A.R. Slabas, P.C. Young)

Chairman: J.E.Godfrey, B.Sc., A.R.Ag.S., gained his degree in agriculture from the University of Reading; he is a director of family farming companies in Lincolnshire and Yorkshire. He is chairman of Sentry Farming Group plc, managing farms in the UK, Poland and the Czech Republic. He is a member or adviser to numerous agricultural committees including The Centre for Agricultural Strategy, University of Reading; The Royal Agricultural Society of England; Food Chain Group of The Foresight Programme; and Humberside Training and Enterprise Council. He is a director of World Potato Congress Inc. He joined the Governing Body of SCRI in 1991, became Vice Chairman in 1998 and Chairman in October 1999.

Professor J.J.F. Belch, M.B., Ch.B., F.R.C.P., M.D., is Professor of Vascular Medicine at the University of Dundee, where she is interested in the causes, manifestations and treatment of disease of the blood vessels and circulation. Additionally she is a member of the Medical Research Council Advisory Board, a member of the Scottish Office Acute Services Review Sub-Committe on Peripheral Arterial Disease, and UK Chairman of the Forum on Angiology. Her interests in terms of crop research relate to the antioxidant content of food, specific fatty acid types within oils, and the relationships of these to vascular disease. She was appointed to the Governing Body of SCRI in 1998.

Professor R.J. Cogdell, B.Sc., Ph.D., F.R.S.E., was awarded his two degrees by Bristol University, and completed his post-doctoral research in the USA. He joined the Botany Department of Glasgow University (now the Institute of Biomedical and Life Sciences) in 1975, and currently holds the Hooker Chair of Botany there. He was awarded a Humbolt Research Prize in 1995. He was appointed to the Governing Body of SCRI in 1997.

J. M. Drysdale is a specialist cereal grower, who farms near Kirkcaldy, Fife, and is Chairman of the Tayforth Marketing Group. He is currently on the Committee of Management of the Scottish Society for Crop Research. He was appointed to the Governing Body of SCRI in 1997.

Professor J. Evans O.B.E., B.Sc., Ph.D., D.Sc., F.I.C.For, is Professor in Tropical Forestry (part time) at Imperial College, London, and was formerly Chief Research Officer (S) with the UK Forestry Commision from 1989 to 1997. He is Vice-Chairman of the Commonwealth Forestry Association and is Chair of DFID's Programme Advisory Committee for Forestry Research. Professor Evans also holds an honorary Chair of Forestry at the University of North Wales, Bangor. He is the author of 7 technical books, including the standard text on tropical forest plantations. Professor Evans owns and manages his own small woodland. He was appointed to the Governing Body of SCRI in 1998.

K. Hopkins, F.C.A., joined Reeves & Neylan, Chartered Accountants, in Canterbury, Kent, in 1971, from a farming background. He moved to open the Scottish Practice in 1978 and was appointed a partner in 1981. "The Scottish Partnership" (a separate business since April 1996) acts for over 500 farmers in Scotland, and specialises in the establishment of farmer-led agricultural cooperatives. His firm now has three offices, Forfar, Perth and Dundee, and employs over 60 staff. Mr Hopkins specialises in capital taxes, agricultural law and cooperatives, development and expansion of business, writes for the agricultural press, and lectures throughout Scotland. He enjoys cricket and golf. He is Treasurer for District 1010 of Rotary and Chairman of the charity Childlink Scotland. He was appointed to the Governing Body of SCRI in 1997.

Professor B. King, M.Sc., Ph.D., F.I.W.Sc., C.Biol., F.I.Biol., is Principal and Vice-Chancellor, University of Abertay Dundee, having joined it in 1992 from the Robert Gordon University, Aberdeen, where he was Assistant Principal and Dean of the Faculty of Health and Food. He is a Non-Executive Trustee of Tayside Primary Care NHS Trust, Board Member of Scottish Enterprise Tayside, Governor of the Unicorn Preservation Society, Chairman of the Conference of Scottish Centrally-Funded Colleges, and Director of the Universities and Colleges Admissions Service (UCAS). He is a member of the International Research Group on Wood Preservation and of the Biodeterioration and British Mycological Societies. He was appointed to the Governing Body of SCRI in 1998.

Emeritus Professor Sir John S. Marsh C.B.E., M.A., P.G. Dip. Ag. Econ., F.R.A.S.E., F.R.Ag.S., C.Biol., F.I.Biol., was Professor of Agricultural Economics, University of Aberdeen, from 1977-1984, then Professor of Agricultural Economics, University of Reading from 1984-1997. He is a Former Director of the Centre of Agricultural Strategy and Chairman of the Agricultural Wages Board, and is currently Chairman of RURAL Council, Governor of the Royal Agricultural College, and Member of the Agriculture, Horticulture and Forestry Foresight Panel. He was made a Knight Bachelor in the Queens Birthday Honours List in 1999 for his wide-ranging contributions to agriculture and agricultural research. He was appointed to the Governing Body of SCRI in 1998.

J.M. Sime, M.Sc., Ph.D., F.R.S.C., C. Chem., is the Chief Executive of the BioIndustry Association, a position he has held since 1995. Prior to this appointment, he held R&D, general management, and strategic marketing positions with Beecham and then SmithKline Beecham, in the UK, USA, Japan, Indonesia, Australia and New Zealand. He is a member of the CBI Biostrategy Committee and of the Management Board of the Advanced Centre for Biochemical Engineering at University College, London. He was appointed to the Governing Body of SCRI in 1997.

Professor A.R. Slabas, B.Sc., D.Phil., is Director of Research, Department of Biological Sciences, University of Durham, where he leads a team involved

in various aspects of lipid metabolism ranging from novel gene identification to structural studies. He has extensive collaboration with Industry, including Biogemma, Zeneca, Linnaeus and Unilever. He is a panel member of the UK Technology Foresight Programme 'Crops for food and Industrial use'; the Eukaryotic Cell Link Management Committee; and the BBSRC Inovative Manufacturing Committee. He joined the Governing Body in 1995

P. Whitworth, H.N.C., retired from United Biscuits as Technical Director, Snacks in March 1996. He has been associated with the production of potato crisps and savoury snacks for over 35 years. He joined the board of the European Snacks Association (ESA) in 1988, and served as President of the Association from 1994 to 1996. He was a founder member of the Board of ECSA Research Ltd (ERL) (the research company formed by ESA to progress the industry's ECLAIR project to improve the tolerance of potatoes to low temperature sweetening using genetic manipulation. Part of this ECLAIR project has been carried out at SCRI.). He has now retired from the board of ERL. He was appointed to the Governing Body of SCRI in 1997.

Professor P.C. Young, B.Tech., M.Sc., M.A., Ph.D., Wh.F., C.Eng., M.I.E.E., F.I.M.A., F.R.S.S., is Director of the Centre for Research on Environmental Systems and Statistics, Lancaster University. He was Head of the Environmental Science Department at Lancaster, 1981-87; Professorial Fellow at the Australian National University, Canberra 1975-81; and Lecturer in Engineering/Fellow of Clare Hall, Cambridge University, 1970-75. His main research interests are in mathematical modelling, time series analysis, forecasting and automatic control. He has worked in a wide range of application areas but his research on agricultural systems includes modelling and advanced control of the micro-climate in horticultural glasshouses; and the data-based mechanistic modelling of biological, horticultural and ecological systems. He is a member of the Board of the Silsoe Research Centre and has been a Member of the Council, Freshwater Biological Association. He was appointed to the Governing Body of SCRI in 1997.

Staff list as at 31 December 1998

Director	Professor J.R. Hillman, B.Sc., Ph.D., D.Sc., F.L.S., C.Biol., F.I.Biol., F.I.Hort., F.R.S.E.[1,2,3,13]	Band 1
Deputy Director	Professor T.M.A. Wilson, B.Sc., Ph.D., C.Biol., F.I. Biol.[2]	Band 2
Secretary & Financial Manager	R.J. Killick, B.Sc., M.B.A., M.A., Ph.D., C.Biol., M.I.Biol.	Band 3
Assistant to Director	T.J.W. Alphey, B.Sc., Ph.D., C.Biol., M.I.Biol.	Band 4

Cell & Molecular Genetics Department (CMG)

Acting Head : G.C. Machray, B.Sc., Ph.D.[1]	Band 4
J.W.S. Brown, B.Sc., Ph.D.[6]	Band 4
R. Ellis, B.Sc., Ph.D.[6]	Band 4
B.P. Forster, B.Sc., Ph.D.[6]	Band 4
W.T.B. Thomas, B.Sc., Ph.D.	Band 4
C.G. Simpson, B.Sc., Ph.D.	Band 5
J.S. Swanston, B.Sc., Ph.D., C.Biol., M.I.Biol.	Band 5
Gillian Clark, H.N.C., B.Sc.	Band 7
B. Harrower, H.N.D., B.Sc., M.Sc.	Band 7
Jackie Lyon	Band 7
Clare McQuade, B.Sc.	Band 7
G.R. Young, H.N.C.	Band 7
A. Booth, H.N.C.	Band 8
Diane Davidson	Band 8 (P/T)
R. Keith	Band 8
M. Macaulay, H.N.C., B.Sc.	Band 8
A. Wilson	Band 8
J.D. Fuller	Band 9
Patricia E. Lawrence	Band 9
Jennifer Watters, H.N.D.	Band 9 (P/T)
Alice Bertie	Band 10
M. Burton	Band 11(P/T)
W. Powell, B.Sc., M.Sc., Ph.D., D.Sc.[4,5,10]	Band 3 (on secondment)

Cellular & Environmental Physiology Department (CEP)

Head : H.V. Davies, B.Sc., Ph.D., C.Biol., F.I.Biol[5,14]	Band 3
J.W. Crawford, B.Sc., Ph.D.[7,15]	Band 4
B.S. Griffiths, B.Sc., Ph.D.	Band 4
Linda L. Handley, B.A., B.Ed., M.Sc., Ph.D.[8]	Band 4
D.K.L. MacKerron, B.Sc., Ph.D.	Band 4
B. Marshall, B.Sc., A.R.C.S., Ph.D.[7]	Band 4
I.M. Morrison, B.Sc., Ph.D.[6]	Band 4
K. Ritz, B.Sc., Ph.D.[7,16]	Band 4
D. Robinson, B.Sc., Ph.D.[6]	Band 4
G.R. Squire, B.A., Ph.D.	Band 4
M.A. Taylor, B.Sc., Ph.D.[9]	Band 4 (Prom. Jul)
R. Viola, B.Sc., Ph.D.	Band 4
R.E. Wheatley, B.Sc., Ph.D.	Band 4 (Prom. Jul)
I.M. Young, B.Sc., Ph.D.[6]	Band 4
A.G. Bengough, B.Sc., Ph.D.	Band 5
N. Deighton, B.Sc., Ph.D., C.Chem., M.R.S.C.	Band 5
J. Liu, B.Sc., M.Sc., Ph.D.	Band 5 (Prom. Jul)
G.J. McDougall, B.Sc., Ph.D.	Band 5
D. Stewart, B.Sc., Ph.D.	Band 5
Sheila Glidewell, M.A., M.Sc., Ph.D.	Band 6
D.C. Gordon, H.N.C.	Band 6
P.D. Hallett, B.Sc., Ph.D.	Band 6
Heather A. Ross, H.N.C., Ph.D., C.Biol., M.I. Biol.	Band 6
Sandra Caul, H.N.C.	Band 7
Susan Verrall, H.N.C.	Band 7
Gladys Wright, H.N.C.	Band 7
D. Crabb	Band 8
G. Dunlop, O.N.C.	Band 8
Lesley George, H.N.C.	Band 8
Diane McRae, O.N.C.	Band 8
Julie A. Duncan	Band 10 (P/T)
B. McGill	Band 11 (P/T) (HELM)

Chemistry Department (Chem)

Head : W.W. Christie, B.Sc., Ph.D., D.Sc., C.Chem., F.R.S.C.	Band 3
B.A. Goodman, B.Sc., Ph.D., C.Chem., F.R.S.C.	Band 4
D.W. Griffiths, M.A., Ph.D, C. Chem., M.R.S.C.	Band 4 (Prom. Jul)
G.W. Robertson, B.Sc., C.Chem., M.R.S.C.	Band 5
C.M. Scrimgeour, B.Sc., Ph.D.[6]	Band 5
H. Bain, H.N.C., L.R.S.C.	Band 6
G. Dobson, B.Sc., Ph.D.	Band 6
Winifred M. Stein, H.N.C., B.Sc.	Band 6
Fiona Falconer, H.N.C.	Band 8
Jean Wilkie	Band 10
Quality Assurance Officer : T. Shepherd, B.Sc., Ph.D.	Band 6

Crop Genetics Department (CG)

Head : G.R. Mackay, B.Sc., M.Sc., C.Biol., F.I.Biol.[4,5]	Band 3
J.E. Bradshaw, M.A., M.Sc., Ph.D.[6]	Band 4
M.F.B. Dale, B.Sc., Ph.D.[6]	Band 4
R. Waugh, B.Sc., Ph.D.[6]	Band 4
G. Bryan, B.Sc., M.Sc., Ph.D.	Band 5
I. Chapman, B.Sc.	Band 5
W. De Jong, B.Sc., Ph.D.	Band 5
M.J. De,Maine, B.Sc., M.Phil.	Band 5
S. Millam, B.Sc., Ph.D.[6]	Band 5
G. Ramsay, B.Sc., Ph.D.	Band 5
Alison K. Lees, B.Sc., Ph.D.	Band 6
Ruth M. Solomon-Blackburn, B.A., M.Sc.	Band 6
Helen E. Stewart, C.Biol., M.I.Biol.	Band 6
Jane Davidson, B.Sc.	Band 7
Jane McNicoll, H.N.C., B.Sc.	Band 7
G.E.L. Swan	Band 7
D. Todd, B.Sc.	Band 7
R.N. Wilson, N.C.H.	Band 7
Nicky Bonar, H.N.C.	Band 8
M.P.L. Campbell	Band 8
P. Davie, O.N.C.	Band 8
Mairi J. Nicolson, B.Sc.	Band 8 (Appt. Dec)
Moira Myles, O.N.C.	Band 9
Sharon Neilson	Band 9
A. Margaret McInroy	Band 10
Gail Simpson	Band 10

[1] Visiting Professor in the University of Strathclyde
[2] Visiting Professor in the University of Dundee
[3] Visiting Professor in the University of Edinburgh
[4] Honorary Senior Lecturer in the University of St. Andrews
[5] Honorary Senior Lecturer in the University of Dundee
[6] Honorary Lecturer in the University of Dundee
[7] Honorary Research Fellow in the University of Dundee
[8] Honorary Professor of Botany, Florida International University
[9] Honorary Lecturer in the University of Glasgow
[10] Honorary Professor, Oregon State University
[11] Honorary Fellow in the University of Edinburgh
[12] Honorary Lecturer in the University of Aberdeen
[13] Visiting Professor in the University of Glasgow
[14] Professor, Universities of Cordoba and Malaga
[15] Visiting Fellow, University of Abertay Dundee
[16] Visiting Professor, University of Kyoto, Japan
[17] Visiting Professor, University of Zhejiang, China

Fungal and Bacterial Plant Pathology Department (FBPP)

Head : J.M. Duncan, B.Sc., Ph.D.[5]	Band 3
G.D. Lyon, B.Sc., M.Sc., Ph.D., D.I.C.[6]	Band 4
A.C. Newton, B.Sc., Ph.D.[6]	Band 4
P. Birch, B.Sc., Ph.D.	Band 6
D. Cooke, B. Sc., Ph.D.	Band 6
Lizbeth J. Hyman, B.A., M.Sc.	Band 6
R. Lowe	Band 6
I. Toth, B.Sc., Ph.D.[12]	Band 6
Jacqueline Heilbronn, H.N.C., B.Sc.	Band 7
Naomi A. Williams, H.N.C.	Band 7

Nematology Department (Nem)

Head : D.L. Trudgill, B.Sc., Ph.D., C.Biol., F.I.Biol., F.S.O.N.[5]	Band 3
B. Boag, B.Sc., Ph.D.[6]	Band 4
D.J.F. Brown, B.A., Ph.D., C.Biol., F.I. Biol., F.R.S.N., F.S.O.N.[6,17]	Band 4
M.S. Phillips, B.Sc.	Band 4
Vivian Blok, B.Sc., M.Sc., Ph.D.	Band 5
A. Kumar, B.Sc., Ph.D.	Band 5
J.T. Jones, B.Sc., Ph.D.	Band 6
R. Neilson, H.N.C., M.Sc.	Band 6 (Prom. Jul)
Ailsa Smith, B.Sc.	Band 7 (P/T)
Sheena S. Lamond	Band 8
Anne M. Holt	Band 8 (P/T)
Alison Paterson	Band 10 (P/T)

Soft Fruit & Perennial Crops Department (SFPC)

Head : R.J. McNicol, B.Sc.[5]	Band 3
A.T. Jones, B.Sc., Ph.D.[5]	Band 3 (IMP)
A.N.E. Birch, B.Sc., Ph.D., C.Biol., M.I.Biol., F.R.E.S.	Band 4 (Prom. Jul)
R.M. Brennan, B.Sc., Ph.D.	Band 4 (Prom. Jul)
B. Williamson, B.Sc., M.Sc., Ph.D., D.Sc.[6]	Band 4
J.A.T. Woodford, B.A., M.A., Ph.D., F.R.E.S.[6]	Band 4
B. Fenton, B.Sc., Ph.D., C.Biol., M.I.Biol.	Band 5
S.C. Gordon, H.N.C.	Band 5
Julie Graham, B.Sc., Ph.D.	Band 5
G. Thow, B.Sc., Ph.D.	Band 6
Alison Dolan, H.N.C.	Band 7 (P/T))
Gaynor Malloch, D.C.R., B.Sc.	Band 7
Wendy J. McGavin, B.Sc.	Band 7
Sandra L. Gordon, H.N.C.	Band 8
Kay Smith, Dip. H.E.	Band 8
A.W. Mills	Band 8
I. Fleming	Band 10
J. Mason	Band 10
Departmental Administrator : Maureen Murray	Band 8

Virology Department (Vir)

Head : P.F. Palukaitis, B.Sc., Ph.D.[5]	Band 3
M.A. Mayo, B.Sc., Ph.D., C.Biol., M.I.Biol.[5]	Band 3 (IMP)
K.J. Oparka, B.Sc., Ph.D.[5]	Band 3 (IMP)
H. Barker, B.Sc., Ph.D.	Band 4
J.M.S. Forrest, B.Sc., Ph.D.	Band 4
I.M. Roberts, H.N.C., Dip.R.M.S.	Band 4
D.J. Robinson, M.A., Ph.D.[6,17]	Band 4
M. Taliansky, Ph.D., D.Sc.	Band 4 (Prom. Jul)
Lesley Torrance, B.Sc., Ph.D.[6]	Band 4
G.H. Duncan, H.N.C.	Band 5
S.A. MacFarlane, B.Sc., D.Phil.	Band 5
B. Reavy, B.Sc., D.Phil.	Band 5
S. Santa Cruz, B.Sc., Ph.D.	Band 5
Jelena Andrejeva, B.Sc., M.Sc., Ph.D.	Band 6 (Appt. Aug)
T. Canto, B.Sc., Ph.D.	Band 6
Maud M. Swanson, B.Sc., Ph.D.	Band 6
Kathryn M. Wright, M.A., Ph.D.	Band 6
A. Ziegler, B.Sc., Ph.D.	Band 6
G.H. Cowan, H.N.D., M.Sc.	Band 7
Sheila M.S. Dawson, H.C.	Band 7
Kara D. McGeachy, H.N.C.	Band 7
Jill Middlefell-Williams, H.N.C.	Band 7
Fiona Carr	Band 8
Gillian L. Fraser	Band 8 (P/T)

Scientific Liaison & Information Services Department (SLIS)

Head : W.H. Macfarlane Smith, B.Sc., Ph.D., C.Biol., M.I.Biol., F.I. Mgt.	Band 4
D.F. Marshall, B.Sc., Ph.D.	Band 4
T.G. Geoghegan, A.B.I.P.P., A.M.P.A.	Band 5
T.D. Heilbronn, B.Sc., M.Sc.	Band 6
I.R. Pitkethly, H.N.D.	Band 6
P. Smith, B.Sc.	Band 6
Sarah E. Stephens, B.Sc., M.A., A.L.A.	Band 6
I. Black, H.N.C.	Band 7
S. Clark, H.N.C., M.Sc.	Band 7
S.F. Malecki, A.B.I.P.P.	Band 7
Ursula M. McKean, M.A., Dip. Lib.	Band 7
G. Menzies	Band 7
Janette Keith	Band 10
Safety Coordinator : M.J. De,Maine, B.Sc., M.Phil.	Band 5

Administration Department (Admin)

Secretary & Financial Manager : R.J. Killick, B.Sc., M.B.A., M.A., Ph.D., C.Biol., M.I.Biol.	Band 3
Financial Controller : I.F. Harrington, C.A.	Band 4
Assistant Secretary : D.L. Hood, B.Admin., Dip. Ed., L.T.I., A.I.I.M.	Band 6
Personnel Officer : I. Paxton, H.N.C., M.Sc., M.I.P.D.	Band 6
Anne Pack	Band 7
Catherine Skelly	Band 7
Dianne L. Beharrie, Dip. Ed.	Band 8
Joyce Davidson	Band 8
Rhona G. Davidson	Band 8
Pam Duncan	Band 8
Sheena Forsyth	Band 8
Kristy L. Grant, B.A.	Band 8
Barbara V. Gunn	Band 8
Theresa Ower, B.A.	Band 8
Sarah-Jane Simms, H.N.D.	Band 8
Elizabeth L. Stewart	Band 8
Sharon Inglis	Band 9 (Appt. Jan)
Louise Fiddes	Band 10 (Appt. Jun)
Media Kitchen	
Wendy Ridley	Band 7
Evelyn Warden	Band 9
W. Burry	Band 11 (HELM)
M.Burton	Band 11(P/T) (HELM)
J. McMillan	Band 11 (P/T) (HELM)

Engineering & Maintenance Department (EM)

Head: S. Petrie, B.Sc.	Band 4
D. Gray, H.N.C.	Band 6
A. Low	Band 7
I.C. McNaughton, H.N.C.	Band 7
K. Henry	Band 8
R.D. McLean	Band 8
G.C. Roberts	Band 8
R. White	Band 8
J. Anderson	Band 9
D. Byrne	Band 9
E. Lawrence	Band 9
C.G. Milne	Band 9
R. Pugh	Band 9
W. Scott	Band 9
C. Conejo	Band 10
J. Flight	Band 10
N. McInroy	Band 10
D.L.K. Robertson	Band 10
J. Rowe	Band 10
M.J. Soutar	Band 10
J. Oldershaw	Band 11
Departmental Administrator :	
Wendy A. Patterson, H.N.D.	Band 8 (P/T)

Estate, Glasshouse & Field Research Department (EGFR)

Head : G. Wood, B.Sc., Ph.D., F.E.T.C.	Band 4
P.A. Gill, H.N.D., N.E.B.O.S.H.	Band 5 (Prom. Jul)
J.R.K. Bennett	Band 7
W.D.J. Jack, B.Sc.	Band 7
D.S. Petrie	Band 7
B.D. Robertson, N.E.B.S.M., H.N.C., Dip. Mgt., M.B.A.	Band 7
R. Ogg	Band 8
D.G. Pugh	Band 8
Angela M. Thain, H.N.C.	Band 8
J.T. Bennett	Band 9
E.R. Caldwell, H.N.C.	Band 9 (Appt. Jun)
B. Fleming	Band 9
L.A. McNicoll	Band 9
Alison Dobson, H.N.C.	Band 10
A.C. Fuller	Band 10
P. Heffell, O.N.C.	Band 10 (Appt. Mar)
T.A. Mason, N.E.B.S.M.	Band 10
M.D. Neill	Band 10 (Appt. Feb)
Gillian Pugh	Band 10
J.K. Wilde	Band 10
J. Abernethy	Band 11 (P/T) (HELM)
M. Torrie	Band 11 (P/T) (HELM)
Departmental Administrator :	
Wendy A. Patterson, H.N.D.	Band 8 (P/T)

Biomathematics and Statistics Scotland (BioSS)

King's Buildings, University of Edinburgh	
Director : R.A. Kempton, M.A., B.Phil.[11]	Band 3
C.A. Glasbey, M.A., Dip. Math. Stats., Ph.D., D.Sc., M.I.S.I.[11]	Band 3 (IMP)
G.J. Gibson, B.Sc., Ph.D.	Band 4
E.A. Hunter, B.Sc., M.Phil.[11]	Band 4
M. Talbot, F.I.S., M.Phil. [11]	Band 4
Janet M. Dickson, B.Sc.	Band 5
I.J. McKendrick, B.Sc., Ph.D.	Band 5
A.M. Roberts, B.Sc., M.Sc.	Band 5 (Appt. Nov)
Elizabeth J. Austin, M.A., D.Phil.	Band 6
J.A.N. Filipe, B.Sc., M.Sc., Ph.D.	Band 6
A.D. Mann, B.Sc.	Band 6
Muriel A.M. Kirkwood, D.A.	Band 8
Karyn Linton	Band 9 (P/T)
Diane Glancy	Band 10 (P/T)
Amy G. Stewart	Band 10 (P/T)
Administration Officer : Elizabeth M. Heyburn, M.A.	Band 7
Ayr Unit	
I.M. Nevison, M.A.	Band 6
Aberdeen Unit, RRI	
Head : G.W. Horgan, B.A., M.Sc., Ph.D.	Band 5
Carol A. Reid, B.Sc., Dip. Acc., Ph.D.	Band 6
Aberdeen Unit, MLURI	
Head : D.A. Elston, B.A., M.Sc.	Band 4
Elizabeth I. Duff, B.Sc.	Band 6
M.E.H. Hodgson, B.A., Ph.D.	Band 6 (Appt. Nov)
Jacqueline M. Potts, B.Sc., M.Sc., Ph.D.	Band 6 (Appt. Oct)
T.S. Smart, B.A., P.G.C.E., M.Sc.	Band 6
Dundee Unit	
Head : J.W. McNicol, B.Sc., M.Sc.	Band 4
Christine A. Hackett, B.A., Dip. Math. Stats., Ph.D.	Band 5
F.G. Wright, B.Sc., M.Sc., Ph.D.	Band 5

Short-term Contracts

SOAEFD Flexible Funding

BioSS

D. Allcroft, B.Sc., M.Sc. — Band 6 (P/T) (Appt. Oct)
Nicole H. Augustin, M.Sc. — Band 6
Maria L. Durban-Reguera, B.Sc., Dip. Math. Stats., Ph.D. — Band 6
Cathryn M. Hau, B.Sc., M.Sc. — Band 6 (P/T) (Appt. Oct)
S.J. Ferris, B.Sc., M.Sc. — Band 7 (Appt. Oct)

Cellular and Environmental Physiology

A.M. Johnston, B.Sc., Ph.D. — Band 5
C. Clegg, B.Sc., M.Sc., Ph.D. — Band 6
Lynda Deeks, B.Sc., Ph.D. — Band 6
P.P.M. Iannetta, B.Sc., Ph.D. — Band 6
Diane Masson, B.Sc., Ph.D. — Band 6 (Appt. Jan)
X. Zhang, B.Sc., Ph.D. — Band 6 (Appt. Jan)
Joanna Chessell, B.Sc. — Band 7
G. Henderson, B.Sc., M.Sc. — Band 7
D. Kiezebrink, B.Sc., M.Sc. — Band 7
M.O. Henry, B.Sc. — Band 7
Fiona C.M. Milne, H.N.D., B.Sc. — Band 7
Evelyn Good — Band 9 (P/T)

Crop Genetics

Karen McLean, B.Sc. — Band 7 (Tr. from CMG May)

Soft Fruit & Perennial Crops

Nicola Wood, B.Sc., Ph.D. — Band 6 (Appt. Sep)
Linzi Ross, H.N.D. — Band 10

Nematology

Lindsey Gray, B.Sc. — Band 6
Lydia Castelli, B.Sc., M.Sc. — Band 7 (Appt. Sep)

Virology

Petra Boevink, B.Sc., Ph.D. — Band 6 (Appt. Oct)
C. Lacomme, B.Sc., Ph.D. — Band 6
Alison Roberts, B.Sc. — Band 6 (Appt. Sep)
S. Wilson — Band 7 (Appt. Feb)

BBSRC

BioSS

Z. Luo, B.Sc., M.Sc., Ph.D. — Band 6 (Appt. Aug)
Grietje Zuur, M.Sc., Ph.D. — Band 6

Cell and Molecular Genetics

Linda Cardle, B.Sc., Ph.D. — Band 6
Julie Wardrop, B.Sc., Ph.D. — Band 6 (Appt. Jul)

Virology

Lisa Smolenska, B.Sc. — Band 7

British Potato Council

Crop Genetics

A.J. Hilton, B.Sc., M.Sc. — Band 6 (Appt. Jul)

Fungal and Bacterial Plant Pathology

Anna Avrova, B.Sc., Ph.D. — Band 6 (Appt. Sep.)
K. Bell, B.Sc., Ph.D. — Band 7
Jane Roberts, H.N.C. — Band 10
Louise Sullivan, B.Sc. — Band 10 (Appt. Jul)

Nematology

M. Elliot — Band 7

CEC

Cell and Molecular Genetics

P. Hedley, B.Sc., Ph.D. — Band 6
L. Ramsay, B.Sc., Ph.D. — Band 6

Cellular and Environmental Physiology

J. Pelloux, B.Sc., Ph.D. — n/a

Chemistry

Claire Fernie, B.Sc. — Band 7

Crop Genetics

D. Milbourne, B.Sc., Ph.D. — Band 6 (Appt. April)
Jane S. Miller, B.Sc., Ph.D. — Band 6
Fiona M. Walls, H.N.D., B.Sc., M.Sc. — Band 8

Fungal and Bacterial Plant Pathology

Alia Dellagi, B.Sc., Ph.D. — Band 6
D.C. Guy, H.N.D. — Band 7

Nematology

S. Pearce, B.Sc., Ph.D. — Band 6 (Appt. Oct)
I. Bendezu, B.Sc., Ph.D. — Band 7 (Appt. Feb)
Alison Prior, B.Sc. — Band 7
Jane Wishart, B.Sc. — Band 7

Soft Fruit & Perennial Crops

I. Muckenschnabel, B.Sc., Ph.D. — Band 6 (Appt. Jul)
R. Weir, B.Sc. — Band 10 (Appt. Mar)

Virology

Rosemary Clarke, B.Sc., Ph.D. — Band 6 (Appt. Dec)
Elaine Ferguson, B.Sc., M.Phil. — Band 6 (Appt. Jan)
Rachel Toth, B.Sc., Ph.D. — Band 6

DFID

Soft Fruit & Perennial Crops

Alyson McRobbie — Band 9 (Appt. Oct)

DTI/LINK

Cell and Molecular Genetics

Adel Ibrahim, B.Sc., Ph.D. — Band 6 (Appt. Sep)
Rhonda Meyer, B.Sc., Ph.D. — Band 6
Jennifer Ritchie, O.N.C. — Band 10

Cellular and Environmental Physiology

N. Nunan, B.Sc., Ph.D. — Band 6 (Appt. Oct)

Virology

Karen Harper, B.Sc., Ph.D. — Band 6

H-GCA

Cell and Molecular Genetics

Elaine Ferguson, B.Sc., M.Phil. — Band 6 (Appt. Jun)

HortLink

Soft Fruit & Perennial Crops

Heather McCafferty, B.Sc., Ph.D. — Band 6
A. Stevenson — Band 10 (Appt. Jun)

MAFF

Cell and Molecular Genetics

A. Ibrahim, B.Sc., Ph.D. — Band 6
Jennifer Watters, H.N.D., B.Sc. — Band 8 (P/T)

Cellular and Environmental Physiology

M. Young, H.N.D., M.Sc. — Band 6
J. Bown, B.Sc, Ph.D. — Band 7 (P/T) (Appt. Oct)
Kirsty Harris, B.Sc. — Band 7 (Appt. Oct)
Sarah Tiller, B.Sc., M.Sc. — Band 7 (Appt. Apr)
Paula M. Hebden, B.Sc. — Band 8

Crop Genetics

Caroline Thompson, B.Sc., Ph.D. — Band 6

Fungal and Bacterial Plant Pathology

D. Cullen, B.Sc., Ph.D. — Band 6

Soft Fruit & Perennial Crops

Trudi Gillespie, B.Sc. — Band 7

Virology

Michele Liney — Band 7

McCains PLC

Crop Genetics

Hayley Baldie — Band 9

Millennium Fund

Irene E. Geoghegan, M.Sc. — Band 7

SmithKline Beecham R&D Fund

Cellular and Environmental Physiology

R.D. Hancock, B.Sc., Ph.D. — Band 6 (Appt. Apr)
M.R. MacLeod, B.Sc., Ph.D. — Band 6
Nicola McCallum, B.Sc. — Band 10 (Appt. Feb)
A. Smith, H.N.C., B.Sc. — Band 10 (Appt. Feb)

Miscellaneous funding

Soft Fruit & Perennial Crops

P. Lanham, B.Sc., Ph.D. — Band 6

Fungal and Bacterial Plant Pathology

K. Holmes, B.Sc., Ph.D. — Band 6 (Appt. May)
Wendy Smith, B.Sc. — Band 7 (Appt. Apr)

Resignations

Name	Dept.	Band	Month
Elizabeth J. Austin	BioSS	6	September
E. Caldwell	EGFR	10	May
A. Grant	EGFR	8	October
D.J. Hirst	BioSS	5	July
S. McDonald	CG	8	October
L. McNicol	EGFR	9	May
D.A. McNulty	BioSS	6	February
T.S. Smart	BioSS	6	April

Staff Retirements

Name	Dept.	Band	Month
Norma Dow	CG	8	January
S.L. Howie	Admin	5	March

Voluntary and Flexible Retirements

Name	Dept.	Band	Month
Karen A. Robertson	BioSS	7	March
W.M. Robertson	Nem	4	Mar
D.A.M. Prior	Vir	7	Feb

Mylnefield Research Services Ltd

Managing Director : N.W. Kerby, B.Sc., Ph.D., C.Biol., F.I.Biol
Commercial Manager : J.B. Snape, M.A., M.Sc., Ph.D. (Appt. Sep)
Administrative Executive Officer : Anne Ross, H.N.C.
Administrative Assistant : Lesley Beaton, H.N.C. (Appt. Oct)
Personal Secretary : Linda Butler

Sharon Canavan
Emily Cobb, H.N.D.
Wendy Craig, B.Sc., Ph.D.
Patricia Dobson
Jane E. Fairlie, O.N.C.
Yuchao Han, B.Sc., M.Sc., Ph.D.
Angela Ingram, B.Sc.
S. Nikki Jennings, B.Sc.
C. Jones, B.Sc.
Susan Mitchell, B.Sc.
A. Mudie, B.Sc.
Jacqueline Murphy, B.Sc., Ph.D.
Claire Reid, B.Sc.
Sheena Rowbottom, O.N.C., H.N.C.
Joanne Russell, B.Sc., Ph.D.
Julie Squires, B.Sc., Ph.D. (Appt. May)
J. Tonberg, B.Sc.
Mary Woodhead, B.Sc., Ph.D.

Honorary Research Professors

Professor P. Broda, M.A., M.Sc., Ph.D., D.Sc., Hon.D.Sc.
Professor H. Griffiths, B.Sc., Ph.D.
Professor F. Gunstone, B.Sc., Ph.D., D.Sc., F.R.S.C., F.T.S.E., C.Chem.
Professor B.D. Harrison, C.B.E., B.Sc., Ph.D., D.Ag.For., F.R.S., F.R.S.E.
Professor N. L. Innes, O.B.E., B.Sc., Ph.D., D.Sc., C.Biol., F.I. Biol., F.R.S.E., F.I. Hort.
Professor P.H. Nye, M.A., B.Sc., F.R.S.
Professor B. Sleeman, B.Sc., Ph.D., D.Sc., C.Math., F.I.M.A., F.R.S.E.
Professor Janet Sprent, O.B.E., B.Sc., D.Sc., Ph.D., A.R.C.S., F.R.S.E.
Professor Sir W. Stewart, B.Sc., Ph.D., D.Sc., C.Biol., F.I.Biol., F.R.S., F.R.S.E.
Professor C.E. Taylor, C.B.E., B.Sc., Ph.D., F.R.S.E., C.Biol., F.I.Biol.

Honorary Research Fellows

R.A. Brown, B.Sc., M.Sc., Ph.D.
J.G. Harrison, B.Sc., Ph.D.
R.J. Jarvis, M.A., D.Phil.
H.M. Lawson, B.Sc., M.Agr.Sc., Dip.Agric., F.I.Hort.
J. McColl, M.B.E., S.H.M., N.D.H., S.D.H.
A.F. Murant, B.Sc., A.R.C.S., Ph.D., D.I.C., C.Biol., F.I.Biol., F.R.S.E.
M.C.M. Pérombelon, M.B.E., B.Sc., M.Sc., Ph.D.
D.A. Perry, B.Sc., Ph.D.
W.M. Robertson, H.N.C., F.L.S.
P.D. Smith, B.Sc., Ph.D., C.Math., F.I.M.A.

Postgraduate Students

Name	Dept.	Subject
D.J. Allcroft	BioSS	Mathematical modelling of short-term behaviour in farm animals.
M. Armstrong	Nem	Molecular heterogeneity in potato cyst nematodes.
Nicole Augustin	BioSS	Statistical spatio-temporal models with applications in vegetation dynamics.
N. Aziz	CMG	Genetic engineering of crops.
Konstantina Boutsika	Nem	Development of molecular diagnostic protocols for detecting "spraing" tobravirus disease of potato and its vector trichodorid nematodes.
J. Bown	CEP	Individual models of ecological systems.
O. Brendel	CEP	^{13}C and genetic variation in native Scots pine.
A. Campbell	BioSS	Inferential tools for stochastic epidemic modelling.
Joanna Chessel	CEP	Reactive transport in soil.
Elaine Davidson	CEP	Isolation and characterisation of new plant-derived mannose-specific lectins and their use in the diagnosis and mechanistic studies of the infection of mammals with a range of bacteria and viruses.
G. Dunlop*	CEP	Linking germination traits of oilseed rape to DNA markers.
S.J. Ferris*	BioSS	The investigation and control of carryover effects in observer perception and recording.
Shahid Hameed	Vir	Properties and diversity of geminiviruses in Pakistan.
G. Henderson	CEP	Modelling soil-water/structure functions.
Sonia N. Humphris	CEP	Biological control as part of an environmentally friendly future for the eradication of dry rot from buildings.
Edwige Isidore	CG	Construction of an ultra high density linkage map of potatoes.
V. Ivandic	CMG	Simple sequence repeats in relation to adaptation in barley.
C. Jones	CEP	Molecular basis of ripening in *Rubus.*
Irene Karanastasi	Nem	Plant virus sequences involved in particle assembly and transmission by nematodes.
D. Kiezebrink	CEP	Modelling soil and water structure functions to assess the efficiency of pesticides in agricultural soils against plant-pathogenic nematodes.
P. Lava Kumar	SFPC	Assessment of the genetic variation within and between populations of *Aceria cajani*, the mite vector of the agent of sterility mosaic of pigeonpea in different regions of Asia.
S.G. Lane	VIR	Studies on recombinant antibodies to water pollutants.
Fevronia Lioliopoulou	Vir	Studies on molecular interactions between PMTV and its vector, *Spongospora subterranea* f.sp. *subterranea*.
Lucy Mackinnon	CG	Transformation of hemp – a multi-purpose fibre crop.
Gaynor Malloch*	SFPC	Genetic variation in the family Byturidae.
Milena Maule	BioSS	Stochastic modelling in plant epidemiology and ecology.
Hazel McGovern	CEP	The influence of soil biota on soil structural conditions.
R. Neilson*	Nem	The rôle of soil fauna in nutrient cycling as indicated by stable isotopic analysis.
Rebecca Nsubuga	BioSS	Statistical study of the epidemiology of *E. coli* O157 infection in cattle.
Elizaveta Pachepsky	CEP	Modelling phenotypic and genotypic interaction in species-rich grassland.
Barnaly Pande	CG	Linkage mapping in 4x potatoes.
Ederlinda Pascual	Chem	Oxidation processes in coffee.
A.A.F.L.K. Perera	CMG	Molecular diversity in coconut.
Alexandra Popovich	SFPC	Development of a rapid screening system for gene function.
Alison Prior	NEM	Functional characterisation of a secreted protein from potato cyst nematode, *Globodera pallida.*
A. Richardson	CEP	Coniferyl alcohol oxidases in lignifying tissues of higher plants.
Alison Roberts	CEP	Plasmodesmata and virus transport.
Caroline D. Robinson	BioSS	Bayesian methods for segmenting X-ray CT images of sheep.
Louise Shepherd	CEP	Production of novel starches in potato.
Geetha Shilvanth	SFPC	Enhancement of resistance to *Botrytis* grey mould of chickpea using PGIP genes.
Lisa Smolenska	Vir	The use of potato virus X for high level production of foreign proteins in plants.
Edwige Souleyre	CEP	Carbohydrate metabolism during ripening in the fruit of strawberry.
Nicole Soranzo	CMG	Molecular ecology of Scots pine.
Kiri Stanley	SFPC	Towards an understanding of the molecular mechanisms of lectin toxicity to aphids through gut glycoprotein interactions.
K. Stewart	FBPP	Breakdown of *Mlo* resistance under stress.
D. Todd*	CG	The genetic effects and consequences of selection for processing potential in the early generations of a potato breeding programme.
N. Vassilakos	Vir	Genetic determinants of complimentarity and exclusivity of vector transmission of tobraviruses.
E. Vellios	Nem	Molecular elucidation of interaction between plant tobravirus gene products and virus-vector trichodorid nematodes.
Jane Wishart	NEM	Characterisation of *Meloidogyne* species using molecular and immunological techniques.
C-P. Witte	CEP	Modification of urea metabolism in transgenic potato.
Joanna C. Wood	BioSS	Mathematical modelling of *E. coli* infection.
C. Zhang	CMG	Improvement of Chinese wheat cultivars.

* Permanent member of staff

Service on External Committees or Organisations

Name	Position	Committee or Organisation
T.J.W. Alphey	Secretary	Committee of Heads of Agricultural and Biological Organisations in Scotland
	Secretary	Scottish Management Advisory Committee
H. Barker	Member	Association of Applied Biologists (Virology Group)
A.G. Bengough	Committee Member	Scottish Soils Discussion Group
	Joint Coordinator	British Soil Water Physics Group
A.N.E. Birch	Convenor	IOBC Working Group - Host Plant Resistance to Pests
	Member	IOBC Working Group – Genetically Modified Organisms (Global Organising Committee)
Vivian Blok	Committee Member	AAB Nematology Sub-Group
B. Boag	Member	UK National Committee for Biodiversity
	Organising Committee	24th International Nematology Symposium, Dundee
R. Brennan	Adviser	SmithKline Beecham Blackcurrant R&D Committee
D.J.F. Brown	Co-Chairman	Russian Society of Nematology International Symposium
	Member	American Society of Nematology Ad Hoc Committee, International Federation of Nematology Societies
	Member	European & Mediterranean Plant Protection Organization Ad Hoc Committee. *Xiphinema americanum* group nematodes
	Organising Committee	24th International Nematology Symposium, Dundee
J.W.S. Brown	Advisory Board	The Plant Journal
W.W. Christie	Member	Steering Committee, 23rd Congress of the International Society for Fats Research, Brighton 1999
	Member	Committee of the European Section of the American Oil Chemists' Society
	Member	EU Concerted Action Committee
D.E.L.Cooke	Council Member	British Society for Plant Pathology
	Chairman	Phytophthora Committee International Society of Plant Pathology
J.W. Crawford	Member	BBSRC EBS Committee
	Member	CHABOS Biodiversity Working Group
M.F.B. Dale	Member	AAB Plant Breeding Committee
H.V. Davies	Member	EU Scientific Committee for Plants
G. Dobson	Treasurer	Royal Society of Chemistry Lipid Chemistry Group
R.P. Ellis	Member	BSPB Cereal Crop Group
	BSPB Representative	Scottish Colleges Cereals Recommended List Consultative Committee
D.A. Elston	Member	ITE Biometrics Network
	Member	RSS Highland Local Group Committee
B.P. Forster	External Examiner	M.Phil. Course in Plant Breeding and Crop Improvement, University of Reading
	Co-ordinator	Chromosome 4. International Barley Chromosome Mapping
	Member	Management Committee, European COST 824 Action 'Gametic Embryogenesis'
Irene Geoghegan	Member	Dundee Science Centre Committee
	Organising Committee	24th International Nematology Symposium, Dundee
G.J. Gibson	Organising Committee	1st IMA Conference on Mathematics in Communication
C.A. Glasbey	Member	EPSRC Mathematics College
	Member	Council of Royal Statistical Society
	Member	RSS Research Committee
	Chairman	RSS Edinburgh Local Group Committee
	Member	Mathematical Sciences Committee of British Association for the Advancement of Science
	Member	Programme Committee for Workshop on Spatial Models, Inference and Algorithms in Image Analysis, Gothenburg, April 1999
B.A. Goodman	Member	Organising Committee of the Crocon Network, funded by the EU under the ALFA Programme for cooperation with South American countries in "Scientific and Technological Training".
B. Griffiths	Organising Committee	24th International Nematology Symposium, Dundee
T.D. Heilbronn	Finance / Publicity Officer	Association for Crop Protection in Northern Britain
J.R. Hillman	Chairman	SCRI/SASA/SAC Liaison Group
	Chairman	Tayside Biocentre Group
	Deputy Chairman	Board of Directors, Mylnefield Research Services Ltd
	Member	Committee of Heads of Agricultural and Biological Organisations in Scotland
	Member	SOAEFD Joint Consultative Committee for Management Board
	Member	ECRR Board of Management
	Member	SNSA Adviser to Committee
	Member	Senate, University of Dundee
	Member	University of Strathclyde Sub-Board for the Degree of B.Sc. in Horticulture
	Member	Tayside Economic Forum
	Member	PSRE Network Steering Committee
	Member	Perth & Kinross Agricultural Forum
	Member	Board of Directors, BioIndustry Association
	Adviser	International Foundation for Science, Stockholm
D.L. Hood	Secretary & Treasurer	Scottish Society for Crop Research
G.W. Horgan	Committee Member	Royal Statistical Society, Local Group
E.A. Hunter	Member	RSS (Edinburgh) Local Group
J. Jones	Organising Committee	24th International Nematology Symposium, Dundee
R.A. Kempton	Past President	British Region, International Biometric Society
	Member	International Biometric Society Awards Fund Committee
	Chairman	Organising Committee. International Biometric Society - British Region, 50th Anniversary Conference, Edinburgh
	Member	Executive Committee, Edinburgh Centre for Rural Research
	Member	Review Group, National Institute for Biological Standards and Control
	Member	Advisory Committee, M.Sc. in Biological Computation, University of York
	Examiner	M.Sc. in Applied Statistics, University of Oxford
R.J. Killick	Member	Scottish Management Advisory Committee

Name	Position	Committee or Organisation
	Member	BBSRC Pay Advisory Group and Pay Negotiation Team
	Company Secretary	Mylnefield Research Services Ltd
Sheena Lamond	Organising Committee	24th International Nematology Symposium, Dundee
W.H. Macfarlane Smith	Member	BBSRC Joint Committee on Health & Safety
	Member	BSPB Oilseed & Industrial Crops Group
	Member	ECRR PR Officers' Group
	Member	SABRI Safety Officers' Group
	Member	NPTC Plant Variety Development Panel
S. MacFarlane	Member	AAB - Virology Group
G.R. Mackay	Chairman	Steering Committee of Global Initiative on Late Blight
	Chairman	Potato Section of EUCARPIA
	President	EUCARPIA (w.e.f. Oct. 98)
	Member	Users Committee of UK Potato Quarantine Unit (SASA)
D.K.L. MacKerron	Secretary	Potato-Crop-Sub-Committee, SSCR
	Chairman	Potato Crop Network, GCTE Focus 3
	Chairman	Section Physiology, EAPR
	Member	Steering Committee of the Seed Potato Forum
D.F. Marshall	Member	Plant and Animal Genome Analysis (PAGA) Initiative, BBSRC
	Member	Resource Allocation and Stress in Plants (RASP) Initiative, BBSRC
	Chairman	NASC Steering Committee, BBSRC
	Member	Genomics Strategy Review, BBSRC
	Member	Quality of Science Institute Review PMS, BBSRC
	Member	Genome Analysis of Agriculturally Important Traits (GAIT) Initiative, BBSRC
	Member	Agri-Food Directorate Network Group, BBSRC
M.A. Mayo	Chairman	ICTV Plant virus Sub-Committee
	Chairman	ICTV Study Group on Satellite Taxonomy
	Member	ICTV Virus Data Sub-Committee
	Member	Study Groups on luteoviruses, tenuiviruses and unclassified viruses
	Member	IUBS/IUMS International Commission on Bionomenclature
G. McDougall	UK Representative	COST Programme E20 on Wood Fibre Cell Wall Ultrastructure
U.M. McKean	Joint Chair	Scottish Agricultural Librarians' Group
I.M. Morrison	Member	SCI, Agriculture and Environment Group Committee
	Member	COST 814-II Alternative Fibre Crops Group
	Member	Energy and Industrial Cropping Group, National Farmers Union of Scotland
	Member	Organising Committee, AAB Conference on Production and Uses of Starch, Edinburgh
R. Neilson	Organising Committee	24th International Nematology Symposium, Dundee
I. Nevison	Member	UPOV Technical Working Party on Automation and Computer Programs
A.C. Newton	Web Server Manager	British Society for Plant Pathology
	Committee Member	Local Arrangements Committee of the seventh International Congress of Plant Pathology (ICPP98)
	Committee Member	United Kingdom Cereal Pathogen Virulence Survey
	National Delegate	Management Committee of EU Cost Action 817
	Committee Member	Crop Protection in Northern Britain Conference
M.S. Phillips	Organising Committee	24th International Nematology Symposium, Dundee
G. Ramsay	Member	UK Plant Genetic Resources Group
	Member	Users Committee of UK Potato Quarantine Unit (SASA)
B. Reavy	Member	Sequiviridae Study Group of International Committee on Taxonomy of Viruses Plant Virus Sub-committee
K. Ritz	Member	BBSRC Plant & Microbial Sciences Committee Network Group
W. Robertson	Organising Committee	24th International Nematology Symposium, Dundee
D.J. Robinson	Member	DETR Advisory Committee on Releases to the Environment.
G.R. Squire	Chairman	CHABOS Working Group on Vegetation Dynamics
	Project Coordinator	SOAEFD Coordinated Programme in Vegetation Dynamics
	Member	CHABOS Working Group on Environmental Pollution, Biological Remediation and Soil Conservation
S.E. Stephens	Joint Chair	Scottish Agricultural Librarians' Group
	Member	Information Services Group - Scottish Library Association
	Member	Tayside and Fife Library and Information Network
	Working Group Member	British Research Institutes Serials Consortium (BRISC)
	Forum Group Member	Research Councils Library and Information Consortium
M. Talbot	Chairman	Statistics Group of UK Plant Varieties and Seeds Committee
	Member	Statistics Committee of International Seed Testing Association
M. Taliansky	Member	Study Groups of International Committee on Taxonomy of Viruses (Plant Virus Subcommittee) on Umbraviruses and Satellites.
W.T.B. Thomas	Member	COST 817 Action 'Airborne pathogens and cereals'
Lesley Torrance	Member	Management EC COST Action 823 Committee
I.K. Toth	Organising Committee	Crop Protection in Northern Britain 1999
D.L. Trudgill	President	24th International Nematology Symposium, Dundee
R.E. Wheatley	Committee Member	Plant/Microbe Interaction Group, Society for Applied Microbiology
B. Williamson	Treasurer	Association for Crop Protection in Northern Britain
T.M.A. Wilson	Member	Programme Committee, VIIth International Congress of Plant Pathology, Edinburgh
	Co-organiser	Joint Royal Society/RSE Symposium on "A Century of Research"
	Member	Dundee Science Centre Consortium Steering Committee
	Member	Church of Scotland, Society Religion and Technology Project 'Ethics of Genetic Engineering of Non-Human Life'
F.G. Wright	Committee Member	BBSRC Collaborative Computational Project CCP11 in biosequence & structure analysis
	Committee Member	BBSRC/EPSRC Bioinformatics Panel
I.M. Young	Chairman	CHABOS Sub-group 'Stability, Resilience and Function of the Soil Resource'

Short-Term Workers and Visitors

Name	Country of origin	Dept.	Month/yr of arrival	Length of stay
Julia Alexander	UK	CG	Jul 98	2 months
A. Aly	Egypt	CMG	Mar 98	6 months
Jelena Andrejeva	Estonia	Vir	Aug 98	1 year
Pamela Barr	UK	SFPC	Aug 98	1 year
A. Basu	India	CMG	Oct 97	4 months
Pilar Blanco-Camba, Ph.D.	Spain	SFPC	Oct 98	1 year
Kirsten Campbell	UK	CG	Jul 98	10 weeks
Sonia Czarnes	France	CEP	Sep 98	1 year
O. David	France	BioSS	Nov 98	2 weeks
V.V. Dolja	USA	Vir	Jun 98	3 weeks
Isabel M.N.R.D. Duarte	Portugal	Nem	Jul 98	3 weeks
Helge Engelking	Germany	CG	Mar 98	6 months
A. van Esberg	The Netherlands	SFPC	Mar 98	6 months
T. Field	UK	Nem	Jul 98	6 months
Stella Fletcher	UK	CG	Jul 98	10 weeks
K. Forbes	UK	SFPC	Aug 98	1 year
Laura Forsyth	UK	CG	Jul 98	10 weeks
M. Ghosh	India	CMG	Nov 97	4 months
Sarah Goodfellow	UK	CG	Jul 98	10 weeks
P. Gonzalez	Spain	Vir	Sep 98	3 months
S. Gray	USA	Vir	Jul 98	3 months
W.A. Gyamerah	Ghana	Vir	Mar 98	4 months
J. Halliday	UK	CEP	Oct 98	6 months
S. Hangt	Germany	Vir	Jun 98	3 months
N. Harding	UK	CG	Jul 98	3 months
A. Howden	UK	CG	Jun 98	2 months
Sonia Humphris	UK	CEP	Sep 98	5 months
Mairi Hunter	UK	SFPC	Aug 98	1 year
M. Iijima	Japan	CEP	Mar 98	10 months
B.Kh. Iskakov	Kazakhstan	Vir	Jun 98	2 weeks
T. Jackson	UK	BioSS	Sep 98	1 year
Anastasia Korycinsca	UK	SFPC	Sep 98	1 year
D. Li	China	Nem	Aug 98	1 month
R. Lister	UK	Nem	Jan 98	6 months
Alison Lyles	UK	CG	Aug 98	9 months
L. Madden	USA	BioSS	Jun 98	2 months
Jacqueline Marks	UK	CG	Oct 98	6 months
Lynne Meikle	UK	CG	Jul 98	10 weeks
Mansoureh Mirabolfathy	Iran	FBPP	Nov 98	3 months
Mirjana Mlinarevic	Croatia	Nem	Jan 98	3 months
F. Nabugoomu	Uganda	BioSS	Jul 98	1 month
P. Neave	UK	SFPC	Aug 98	1 year
Katrine Nicol	UK	CG	Feb 98	4 months
K. M. Nurkiyanova	Kazakhstan	Vir	Oct 97	2 years
J. Pelloux	France	CEP	Dec 97	15 months
Vlada Peneva	Bulgaria	Nem	Jan 98	3 months
K. Pradel	Germany	Vir	Jan 97	2 years
R. Rabindran	India	Vir	Jan 98	6 months
A. Pedros Rafart	Spain	CEP	Feb 98	6 months
K.L.N. Reddy	India	Vir	May 98	(deceased, Aug 99)
E.V. Ryabov	Russia	Vir	Sep 97	2 years
Jaana Sagulin	Finland	SFPC	Apr 98	4 months
T. Sasaya	Japan	Vir	Oct 98	1 year
G. Sherlock	UK	CG	Jun 98	10 weeks
W. Sledz	Poland	FBPP	Aug 98	2 weeks
P. Solomon	Australia	BioSS	Mar 98	2 months
A. Sopena	Spain	CMG	Jul 98	1 month
D. Szilassy	Hungary	Vir	Feb 98	3 months
R. Taylor	New Zealand	FBPP	Aug 98	1 month
A. Tchernets	Moldova	SFPC	May 98	3 months
M. Terzakis	Greece	SFPC	Jul 98	3 months
Susan Tyler	UK	SFPC	Aug 98	1 year
Irene Walker	UK	SFPC	Aug 98	1 year
Ulrike Wellenreuther	Germany	Vir	Apr 98	2 months
Jacqueline Wilson	UK	CG	Apr 98	1 month
Z. Zhang	China	CMG	Feb 98	1 year
J. Zheng	UK	Nem	Jan 98	3 months

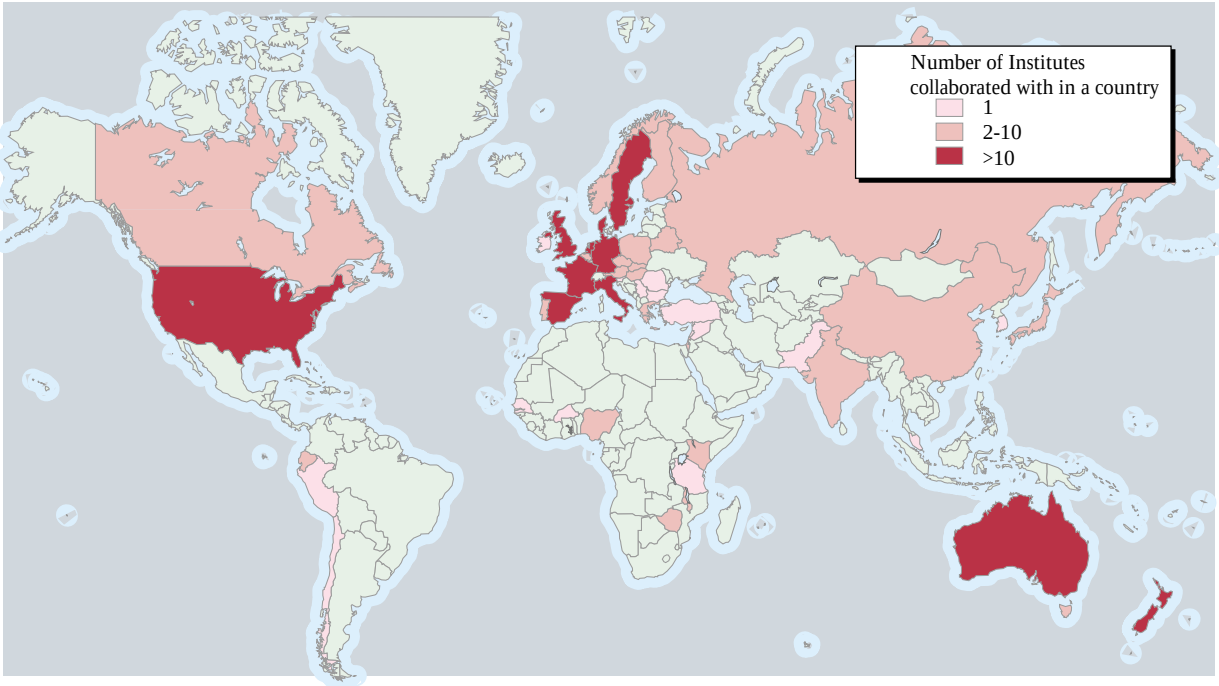
Editorial Duties

Name	Position	Journal Title
H. Barker	Editorial Board	*Annals of Applied Biology*
	Editor	*Descriptions of Plant Viruses on CD ROM*
A.G. Bengough	Editor	*Annals of Botany*
A.N.E. Birch	Editor	*Bulletin of the IOBC (WPRS)*
B. Boag	Editorial Board	*Annals of Applied Biology*
	Editorial Board	*Nematologia Mediterranea*
R. Brennan	Editorial Board	*Journal of Science of Food & Agriculture*
	Associate Editor	*Journal of Horticultural Science & Biotechnology*
D.J.F. Brown	Honorary Chief Editor	*Russian Journal of Nematology*
	Editorial Board	*Nematologia Mediterranea*
	Editorial Board	*Helminthologia*
	Editorial Board	*Journal of Nematode Morphology and Systematics*
W.W. Christie	Editorial Board	*Chemistry and Physics of Lipids*
	Editorial Board	*Lipid Technology*
	Editorial Board	*Grasas y Aceites*
	Editorial Board	*Lipids*
	Managing Editor	The Oily Press Ltd
D.E.L. Cooke	Editor	*Molecular Plant Pathology On-Line*
J.W. Crawford	Editor	*Geoderma*
M.F.B. Dale	Editor	*Annals of Applied Biology*
J.M. Duncan	Associate Editor	*Journal of Horticultural Science & Biotechnology*
G.J. Gibson	Associate Editor	*IMA Journal of Maths Applied in Medicine & Biology*
C.A. Glasbey	Associate Editor	*Biometrics*
	Associate Editor	*Journal of Royal Statistical Society, Series B*
J. Graham	Associate Editor	*Journal of Horticultural Science & Biotechnology*
B.S. Griffiths	Editorial Board	*Pedobiologia*
C. Hackett	Editorial Board	*Heredity*
J.R. Hillman	Publication Committee	*Journal of Horticultural Science*
	Editorial Board	*Agricultural Systems*
	Editorial Board	*Journal of Agricultural Science*
G.W. Horgan	Editor	*British Journal of Nutrition*
E.A. Hunter	Editorial Board	*Food Quality & Preference*
A.T. Jones	Co-Editor	*Descriptions of Plant Viruses, Association of Applied Biologists*
R.A. Kempton	Editorial Board	*Journal of Agricultural, Biological and Environmental Statistics*
D.K.L. MacKerron	Associate Editor	*Journal of Horticultural Science*
	Member of Editorial Board	*Euphytica*
M.A. Mayo	Editorial Board	*Virology*
	Referees Panel	*Nucleic Acids Research*
	Abstractor	*Vitis*
	Editorial Advisory Board	*Encyclopaedia of Life*
	Editorial Board	*Encyclopaedia of Virology*
J.W. McNicol	Editor (Statistical)	*Annals of Applied Biology*
I.M. Morrison	Executive Editor	*Journal of the Science of Food and Agriculture*
R. Neilson	Deputy Chief Editor	*Russian Journal of Nematology*
A.C. Newton	Senior Editor	*Molecular Plant Pathology On-Line*
P. Palukaitis	Senior Editor	*Molecular Plant Microbe Interactions*
	Associate Editor	*Virology*
	Editorial Board	*Journal of General Virology*
M.S. Phillips	Associate Editor	*Journal of Nematology*
K. Ritz	Editorial Board	*FEMS Microbiology Ecology*
D.J. Robinson	Editorial Board	*Journal of Virological Methods*
	Editor	*Molecular Plant Pathology On-Line*
	Editor	*Descriptions of Plant Viruses*
G.R. Squire	Editorial Board	*Experimental Agriculture*
	Advisory Board	*Crop Physiology Abstracts*
D.L. Trudgill	Advisory Board	*European Journal of Plant Pathology*
	Editorial Board	*Nematologica*
	Editorial Board	*Fundamental and Applied Nematology*
	Associate Editor	*Journal of Nematology*
B. Williamson	Editorial Board	*Annals of Applied Biology*
T.M.A. Wilson	Editorial Board	*Journal of General Virology*
F.G. Wright	Member of Editorial Board	*Heredity*
I.M. Young	Associate Editor	*European Journal of Soil Science*
	Associate Editor	*Pedosphere*

Awards and Distinctions

Name	Dept.	Degree/Award/Distinction/Appointment
D.J.F. Brown	Nem	Visiting Professor, Zhejiang University, China
G.C. Machray	CMG	Honorary Visiting Professor, University of Strathclyde
K. Ritz	CEP	Visiting Professor, University of Kyoto, Japan
D.J. Robinson	Vir	Visiting Professor, Zhejiang University, China
A.N.E. Birch	SFPC	Fellow of the Royal Entomological Society
D.J.F. Brown	Nem	Fellow of the Institute of Biology
J.W. Crawford	CEP	Visiting Fellow in Bioinformatics, University of Abertay Dundee
R.M. Brennan	SFPC	University of Minnesota Arboretum Research Chair
I. Muckenschnabel	SFPC	Novartis-Graduierten-Stipendien awarded at Regensburg University
Suzanne Baker	FBPP	Ph.D., University of Oxford
Annette Baty	CEP	Ph.D., University of Dundee
K. Cheung	Nem	Ph.D., University of Leicester
Maria L. Durban-Reguera	BioSS	Ph.D., University of Heriot-Watt
M.E.M. Ehwaeti	Nem	Ph.D., University of Dundee
Liliana F. Franco-Lara	Vir	Ph.D., University of Dundee
J. Hamilton	CMG	Ph.D., University of Dundee
S. Hendy	Vir	Ph.D., University of Leicester
I. Inglis	BioSS	Ph.D., University of Edinburgh
Philabeg Irving	SFPC	Ph.D., University of Dundee
Grainne H. McGuire	BioSS	Ph.D., University of Edinburgh
Anne L. Maddison	CMG	Ph.D., University of Dundee
D. Milbourne	CG	Ph.D., University of Dundee
Adele Mooney	Vir	Ph.D., University of Dundee
Denise Pallett	Vir	Ph.D., University of Dundee
J. Provan	CMG	Ph.D., University of Dundee
L. Robertson	Nem	Ph.D., University of Dundee
Gemma White	CMG	Ph.D., University of Dundee
Grietje Zuur	BioSS	Ph.D., University of Edinburgh
S. Clark	BITR	M.Sc., University of Abertay Dundee
G. Cowan	Vir	M.Sc., University of Dundee
B.E. Harrower	CMG	M.Sc., University of Dundee
Pauline McConway	FBPP	M.Sc., University of Dundee
Jacqueline Heilbronn	FBPP	B.Sc. Hons, University of Dundee
Jennifer A. Watters	CMG	B.Sc., Open University
J.R.K. Bennett	EGFR	D32 Assessor, TDLB
B.D. Robertson	EGFR	D32 Assessor, TDLB
D.G. Pugh	EGFR	D32 Assessor, TDLB
Angela M. Thain	EGFR	D32 Assessor, TDLB
J.T. Bennett	EGFR	SVQ 2, Extensive Crop Production
B. Fleming	EGFR	SVQ 2, Extensive Crop Production

International Collaboration and Consultancies



Research is executed within an international framework that encourages information transfer. The extent of SCRI's international commitment between 1993 and 1998 is reflected in the collaborative research that was undertaken with 318 Institutions in 54 countries. Within the UK, SCRI collaborated with over 200 organisations.

SCRI Research Programme
1998-1999

SOAEFD funded research programme showing: SOAEFD project number; Title (prefixed ROA for ROAMEd core-funded projects; FF for Flexible Fund projects); Scientific Project Leader. In addition to this list, there are research projects undertaken on behalf of various bodies, including other governmental bodies, commerce and levy boards.

SCR/422/94	ROA Processing of plant fibres by novel and environmentally acceptable methods	Davies H V
SCR/424/94	ROA Relating soil structure to biological function	Young I M
SCR/429/94	ROA Genetic architecture of diploid potatoes and production of enhanced germplasm	Bradshaw J E
SCR/432/94	ROA Integrated approaches for rapid and efficient gene transfer and characterisation in potato	Millam S
SCR/444/95	ROA Low temperature stress in *Ribes*, *Rubus* and other woody genera	McNicol R J
SCR/445/95	ROA Collection and evaluation and genetic resources of *Rubus*, *Ribes* and *Fragaria*	McNicol R J
SCR/446/95	ROA Molecular study of genetic variation in plant parasitic nematodes in relation to virulence and plant resistance especially in relation to potato cyst nematodes (PCN) and root knot nematodes	Phillips M S
SCR/449/95	ROA Advanced information techniques for the study and management of vegetation systems	MacKerron D K L
SCR/452/95	ROA Genetic architecture of tetraploid potatoes and production of enhanced germplasm	Bradshaw J E
SCR/454/95	ROA Structure of soil microbial and faunal communities, their interaction with vegetation and the relationship to soil processes and health	Griffiths B S
SCR/455/95	ROA DARE Dynamics and connectivity in discontinuous plant populations, using wild raspberry and feral oilseed rape as model systems	Squire G
SCR/456/95	ROA Genetics and ecophysiology of abiotic stress tolerance in *Hordeum vulgare* (barley) and *Arabidopsis thaliana*	Forster B P
SCR/457/95	ROA Development and evaluation of novel methodology involving modern chromatography and mass spectroscopy for stable isotopes and antinutritional, quality and other biologically active compounds	Christie W W
SCR/462/96	ROA Molecular mechanisms of plant virus replication and movement and the effects of resistance genes on these processes, using cucumoviruses and tobraviruses as contrasting model systems	Palukaitis P F
SCR/464/96	ROA Biochemical and molecular control of carbohydrate metabolism and the modification of starch structure in potato	Davies H V
SCR/465/96	ROA Application and exploitation of molecular markers in barley genetics	Powell W
SCR/471/96	ROA Mathematical analysis of dynamics and scaling in heterogeneous and hierarchically coupled systems I: the soil/microbe complex	Crawford J W

SCR/478/96	ROA Physiological mechanisms underlying the environmental responses of crops in Northern Britain: stable isotope studies of carbon, nitrogen and water relations in barley and contrasting dicot model populations	Handley L L
SCR/479/96	ROA Maintenance, improvement, evaluation and exploitation of biodiversity in germplasm collections of potato	Mackay G R
SCR/481/96	ROA Evaluation, improvement, maintenance and exploitation of biodiversity in germplasm collections of brassicas for improved pest resistance (particularly cabbage and turnip root flies) and nutritional value	Birch A N E
SCR/482/96	ROA Detection, identification, genetic variation and ecology of virus and insect, mite and nematode pests and virus vectors, especially of soft fruit crops, and strategies for their effective control	Jones A T
SCR/483/96	ROA Soft rot erwinias and blackleg disease: aetiology, epidemiology and pathogenicity, selection of resistant potato cultivars and their mechanisms of resistance	Lyon G D
SCR/486/96	ROA Identification and development of control strategies for fungal diseases of fruit crops, especially the use of specific enzyme inhibitors for control of *Botrytis cinerea* in fruit	Williamson B
SCR/487/96	ROA Mathematical analysis of dynamics and scaling in heterogeneous and hierarchially coupled systems II: complex biochemical networks	Crawford J W
SCR/494/97	ROA Genetic control of pathogenicity, host specificity and race structure at the molecular level in the fungal pathogens *Phytophthora infestans, Phytophthora fragariae* and related *Phytophthora* species	Duncan J M
SCR/495/97	ROA Transcriptional and post-transcriptional regulation of plant gene expression	Brown J W S
SCR/496/97	ROA Production of novel diagnostic reagents, in particular genetically engineered antibody-like proteins and investigation of their potential for use in research, biotechnology and diagnosis	Torrance L
SCR/497/97	ROA Studies on mechanisms of host gene-mediated and pathogen-derived transgene-mediated resistance to viruses to improve the deployment of new types of resistance for germplasm enhancement	Barker H
SCR/499/97	ROA Free radical processes in plants and plant-derived foods	Davies H V
SCR/501/97	ROA Develop and operate methods for the detection and quantification of genetic resistance to a wide range of economically important fungal and bacterial pathogens of potato	Bradshaw J E
SCR/508/98	ROA Cell biology of plant-virus interactions	Oparka K J
SCR/509/98	ROA Molecular dissections of plant viral movement proteins	Oparka K J
SCR/510/98	ROA Molecular mechanisms involved in the aphid transmission of luteoviruses, potyviruses and the nematode transmission of tobraviruses	Mayo M A
SCR/511/98	ROA Biological determinants of the specificities of aphid transmission of luteoviruses and of nematode transmission of tobraviruses	Mayo M A
SCR/512/98	ROA Produce and maintain pathogen-tested stocks of *Rubus, Ribes* and *Fragaria* germplasm and index for infection material imported into SCRI	Jones A T

SCR/513/98	ROA Gene expression and manipulation in barley	Machray G C
SCR/514/98	ROA Isolation and functional characterisation of genes important in the host-parasite interaction of plant parasitic nematodes including *Globodera*, *Meloidogyne* and *Xiphinema* spp.	Trudgill D L
SCR/518/98	ROA Towards the construction of a physical and functional map of the interval between GP21 and GP179 on potato linkage group V	Waugh R
SCR/519/98	ROA Characterisation of the *Hero* gene of tomato, which confers resistance to potato cyst nematodes, by transformation into potato	Kumar A
SCR/520/98	ROA Mapping and isolation of *Rhynchosporium secalis* recognition and early signalling genes in barley	Newton A C
SCR/521/98	ROA Genetic modification of soft fruit crops and risk assessment for their introduction	Graham J
SCR/443/95	FF Research into nutritional aspects of genetically manipulated potatoes, *Solanum tuberosum*	Mackay G R
SCR/458/95	FF Determining the origin and genetic structure of late blight outbreaks on Scottish seed and ware potatoes and assessing the hazard of sexual reproduction by *Phytophthora* to the seed industries of Scotland	Duncan J M
SCR/461/95	FF Native Scots Pine: establishing a scientific basis for its conservation	Powell W
SCR/488/96	FF Modelling soil-water/structure functions to assess the efficiency of pesticides in agricultural soils against pathogenic nematodes	Young I M
SCR/504/97	FF Comparison of serological and PCR tests on dormant tubers and attempts to identify sources of virus in Scottish fields	Barker H
SCR/505/97	FF Molecular approaches to manipulate the development and composition of strawberry fruit	Davies H V
SCR/506/97	FF Use of stable isotope techniques to determine the origin, movement and effects of nitrate in the catchment area of the river Ythan	Handley L
SCR/507/97	FF Faecal microbial community structure in VTEC and non-VTEC carrying cattle	Ritz K
SCR/516/97	FF Genetic mapping and molecular cloning of novel sources of resistance to *Globodera pallida*	Waugh R
SCR/522/98	FF Development of *Rubus* genotypes with transgenic resistance to raspberry bushy dwarf virus	Jones A T
SCR/524/98	FF Unravelling the pathways of protein transport in plant and animal cells using virus-based vectors	Oparka K J
SCR/808/94	FF Development of molecular biological and physiological techniques in studies of the interaction between microbes, nutrient cycling and vegetation among a range of agriculturally important pastures, to enable scaling from microcosm to field	Ritz K
SCR/816/95	FF Phenotypic and genotypic bases of population dynamics in heterogeneous, species-rich grassland	Squire G
SCR/818/95	FF Genetic engineering of crop plants for resistance to insect and nematode pests: effects of transgene expression on animal nutrition and the environment	Robertson W M

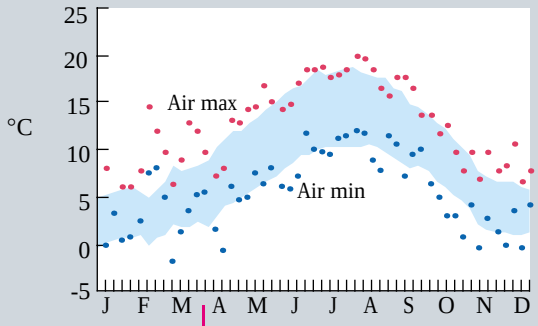
SCR/821/96	FF Exploitation of novel and known lectins in agricultural and biological research - an interdisciplinary approach to improve crop protection and productivity, animal (including human) welfare and health	Stewart D
SCR/822/97	FF The application of the free-living nematode *C. elegans* to the development of control procedures for nematode parasites of animals and plants	Jones J
SCR/823/97	FF Significance of physical heterogeneity for scaling of solute chemistry in soils from fine scale to subcatchment	Crawford J W
SCR/824/97	FF Efficacy studies on a plant virus-based expression system and on alternative delivery routes for peptides and proteins with pharmaceutical, therapeutic and related uses for improving animal health, nutrition and welfare	Wilson T M A
SCR/831/98	FF Insect-resistant transgenic plants in tri-trophic interactions: ecological, biochemical and molecular impact on aphiophagus ladybird predators	A N E Birch

Meteorological Records

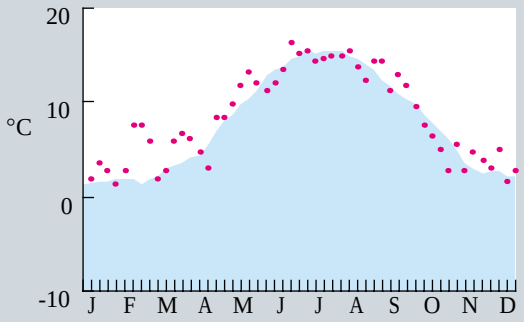
D.K.L. MacKerron

Detailed meteorological records are kept regularly at SCRI. The graphs shown are for weekly values for 1998 and the long term average for 1961-1990( ).

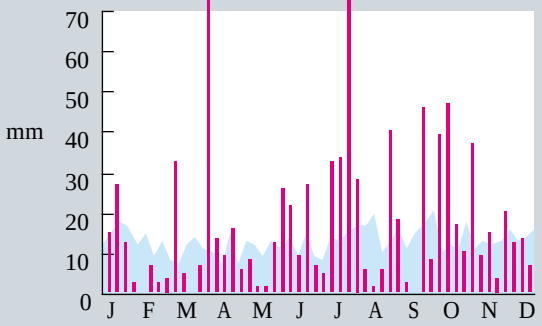
Air temperature



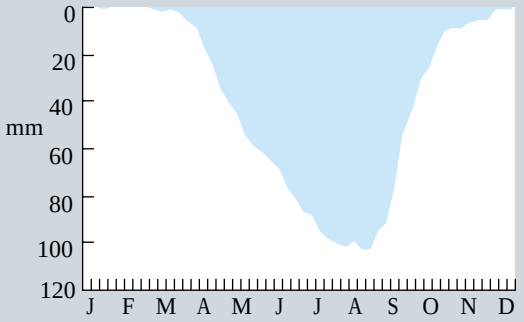
10cm soil temperature



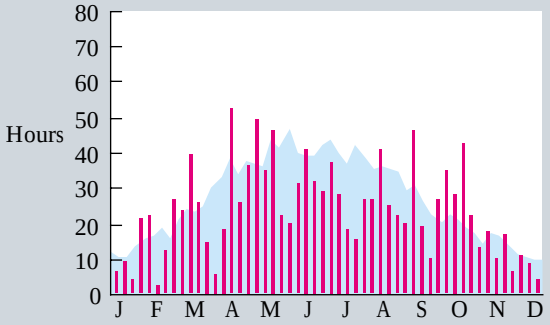
Rainfall



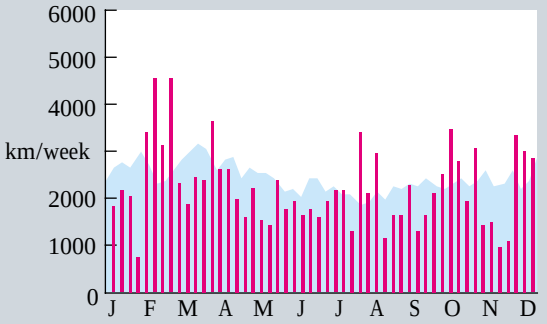
Soil moisture deficit *



Sunshine



Run of wind



* 1998 values for SMD not available

Cumulative Index 1990 - 1998/9

In addition to the list below, in every SCRI Annual Report during this period, there are reports of Mylnefield Research Services Ltd; the Research Services; a General Report including accounts, staff lists, publications, research project lists; Overviews by each Head of Department; and a Report by the Director.

Plant genetics

Breeding and genetics (1997/8 onwards)

Soft fruit and perennial crops

Molecular biology

Plant molecular and cell biology (1997/8 onwards)

Cellular and environmental physiology

Chemistry

Plant biochemistry and phytochemistry (1997/8 onwards)

Fungal and bacterial diseases

Plant viruses

Zoology

Biomathematics and Statistics Scotland

Host pathogen interactions and crop protection

(1997/8 onwards)

Plants, soils and environment (1997/8 onwards)

Other Articles

Institutes supported by the Biotechnology and Biological Sciences Research Council

BBSRC Office	Polaris House, North Star Avenue, Swindon, Wilts SN2 1UH	01793-413200
BBSRC Bioscience IT Services	West Common, Harpenden, Herts AL5 2JE	01582-762271
Babraham Institute	Babraham Hall, Babraham, Cambridge CB2 4AT	01223-832312
Laboratory of Molecular Signalling	Babraham Institute, P.O. Box 158, Cambridge CB2 3ES	01223-336600
Institute for Animal Health	Compton, Near Newbury, Berkshire RG20 7NN	01635-578411
Compton Laboratory	Compton, Near Newbury, Berkshire RG20 7NN	01635-578411
Pirbright Laboratory	Ash Road, Pirbright, Woking, Surrey GU24 0NF	01483-232441
BBSRC & MRC Neuropathogenesis Unit	Ogston Building, West Mains Road, Edinburgh EH9 3JF	0131-667-5204
Institute of Arable Crops Research	Harpenden, Herts AL5 2JQ	01582-763133
IACR - Long Ashton Research Station	Long Ashton, Bristol BS18 9AF	01275-392181
IACR - Rothamsted	Harpenden, Herts AL5 2JQ	01582-763133
IACR - Broom's Barn	Highham, Bury St. Edmunds, Suffolk IP28 6NP	01284-810363
Institute of Food Research	Earley Gate, Whiteknights Rd, Reading RG6 6BZ	01189-357055
Norwich Laboratory	Norwich Research Park, Colney, Norwich NR4 7UA	01603-255000
Reading Laboratory	Earley Gate, Whiteknights Rd, Reading RG6 6BZ	01189-357000
Institute of Grassland and Environmental Research	Plas Gogerddan, Aberystwyth, Dyfed SY23 3EB	01970-828255
Aberystwyth Research Centre	Plas Gogerddan, Aberystwyth, Dyfed SY23 3EB	01970-828255
North Wyke Research Station	Okehampton, Devon EX20 2SB	01837-82558
Bronydd Mawr Research Station	Trecastle, Brecon, Powys LD3 8RD	01874-636480
Trawsgoed Research Farm	Trawsgoed, Aberystwyth, Dyfed SY23 4LL	01974-261615
John Innes Centre	Norwich Research Park, Colney, Norwich NR4 7UH	01603-452571
Roslin Institute	Roslin, Midlothian EH25 9PS	0131-527-4200
Silsoe Research Institute	Wrest Park, Silsoe, Bedford MK45 4HS	01525-860000
Horticultural Research International	Wellesbourne, Warwick CV35 9EF	01789-470382
HRI, East Malling	West Malling, Maidstone, Kent ME19 6BJ	01732-843833
HRI, Wellesbourne	Wellesbourne, Warwick CV35 9EF	01789-470382

Scottish Agricultural and Biological Research Institutes

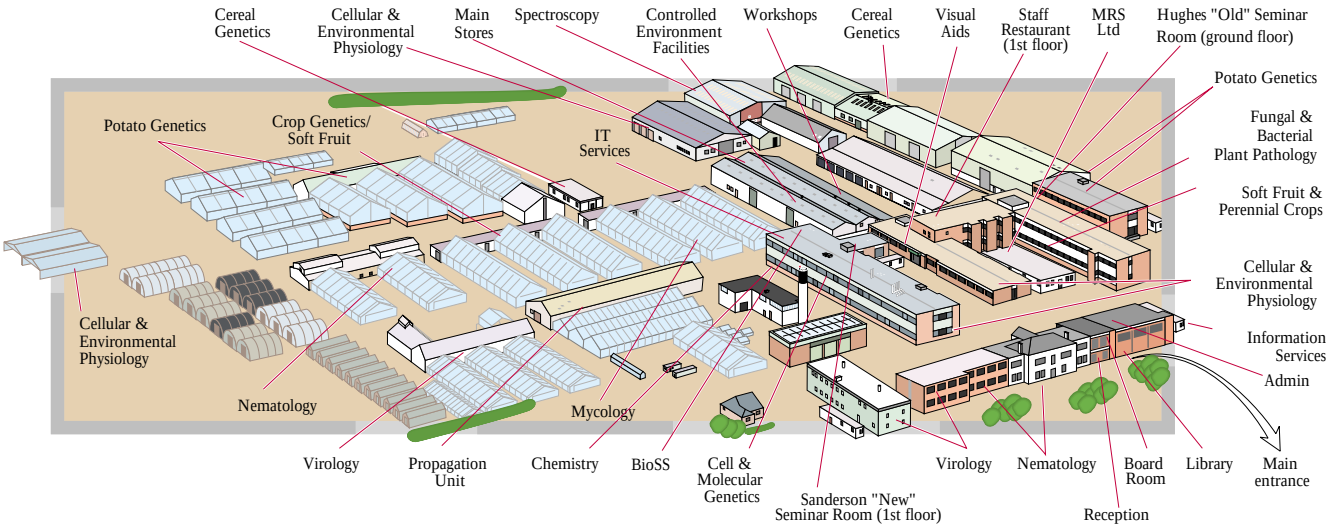
Hannah Research Institute	Ayr, Scotland KA6 5HL	01292-476013
Macaulay Land Use Research Institute	Craigiebuckler, Aberdeen AB9 2QJ	01224-318611
Moredun Research Institute	408 Gilmerton Road, Edinburgh EH17 7JH	0131-664-3262
Rowett Research Institute	Greenburn Road, Bucksburn, Aberdeen AB21 9SB	01224-712751
Scottish Crop Research Institute	Invergowrie, Dundee DD2 5DA	01382-562731
Biomathematics and Statistics Scotland (Administered by SCRI)	University of Edinburgh, James Clerk Maxwell Building, King's Buildings, Mayfield Road, Edinburgh EH9 3JZ	0131-650-4900

List of Abbreviations

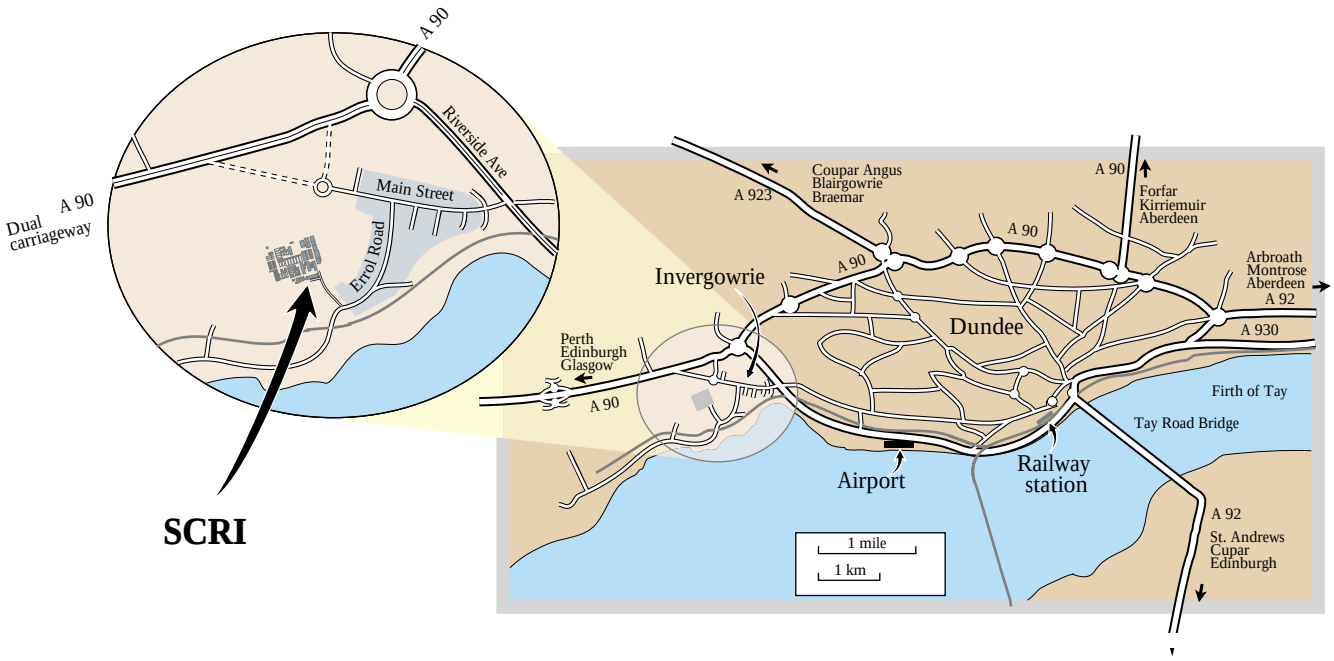
AAB	Association of Applied Biologists
ACRE	Advisory Committee on Releases to the Environment
ADAS	Agricultural Development and Advisory Service
BBSRC	Biotechnology & Biological Sciences Research Council
BCPC	British Crop Protection Council
BioSS	Biomathematics and Statistics Scotland
BPC	British Potato Council
BSPB	British Society of Plant Breeders
BTG	British Technology Group
CAPS	Cleaved Amplified Polymorphic Sequence
CEC	Commission of the European Communities
CHABOS	Committee of Heads of Agricultural and Biological Organisations in Scotland
CIP	International Potato Centre - Peru
COST	European Co-operation in the field of Scientific and Technical Research
DfID	Department for International Development
EAPR	European Association for Potato Research
ECRR	Edinburgh Centre for Rural Research
ECSA	European Chips and Snacks Association
EHF	Experimental Husbandry Farm
ELISA	Enzyme linked immunosorbent assay
EPPO	European Plant Protection Organisation
ESTs	Expressed Sequence Tagged Sites
FF	Flexible Funding (SOAEFD)
FLAIR	Food-Linked Agro-Industrial Research
GILB	Global Initiative on Late Blight
GIUS	Glasshouse Investigational Unit for Scotland
H-GCA	Home-Grown Cereals Authority
HDC	Horticultural Development Council
HPLC	High Performance Liquid Chromatography
HRI	Hannah Research Institute
IACR	Institute of Arable Crops Research
ICTV	International Committee for the Taxonomy of Viruses
IOBC	International Organisation for Biological Control
ISHS	International Society for Horticultural Science
ISPP	International Society for Plant Pathology
IVEM	Institute of Virology and Environmental Microbiology
MAFF	Ministry of Agriculture Fisheries and Food
MLURI	Macaulay Land Use Research Institute
MRI	Moredun Research Institute
NERC	National Environmental Research Council
NFT	National Fruit Trials
NFU	National Farmers Union
NIR	Near Infra-Red
NMR	Nuclear Magnetic Resonance
NPTC	National Proficiency Test Council
ORSTOM	Organisation for research in science and technology overseas
PCR	Polymerase Chain Reaction
PVRO	Plant Variety Rights Office
RAPD	Randomly Amplified Polymorphic DNA
RFLP	Restriction Fragment Length Polymorphism
RRI	Rowett Research Institute
SABRI	Scottish Agricultural and Biological Research Institutes
SAC	Scottish Agricultural College
SASA	Scottish Agricultural Science Agency
SCRI	Scottish Crop Research Institute
SEB	Society for Experimental Biology
SET	Scottish Enterprise Tayside
SNSA	Scottish Nuclear Stocks Association
SOAEFD	Scottish Office Agriculture, Environment and Fisheries Department
SSCR	Scottish Society for Crop Research
SSFG	Scottish Soft Fruit Growers Ltd
STS	Sequence Tagged Sites
UNDP	United Nations Development Programme
WHO	World Health Organisation

The Scottish Crop Research Institute

Site plan



Access to Scottish Crop Research Institute



SCRI is on the east coast of Scotland, midway between Edinburgh and Aberdeen.

It is located at Invergowrie 6km west of the centre of Dundee. Access is via Riverside Avenue, Main Street and Errol Road.

British Rail has direct InterCity services between Dundee and London, Edinburgh and Glasgow and other UK cities.

Flights are available to Dundee Airport from Edinburgh, Manchester and Aberdeen, and scheduled services operate from many domestic and international destinations to Edinburgh and Glasgow.