SCOTTISH PLANT BREEDING STATION

PENTLANDFIELD, ROSLIN, MIDLOTHIAN

REPORT

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STAFF

Scottish Plant Breeding Station

Director	J. W. Gregor, Ph.D., D.Sc., F.R.S.E.
Chief Assistant	W. Black, Ph.D., D.Sc., F.R.S.E.
Brassica Crops—	
Breeding	V. McM. Davey, B.Sc., Ph.D. A. Murray, B.Sc. (*resigned* 31/1/60). *Vacancy.*
Cytogenetics	I. H. McNaughton, B.A., D.Phil. Mrs S. Morrison, S.D.H.
Herbage Plants and Genecology	Miss P. J. Watson, M.A., Ph.D. D. J. Harberd, M.Sc. D. A. Wilkins, B.Sc., Ph.D. F. J. W. England, B.Sc. Miss S. M. Gray, B.Sc.
Oats	D. Cameron, B.Sc. G. R. White, B.Sc. D. W. Speed, B.Sc.
Potatoes—	
Breeding	W. Black. J. M. Dunnett, B.Sc. A. W. Macarthur, B.Sc. Miss J. F. Malcolmson, B.Sc., Ph.D.
Virus Disease Investigations	G. Cockerham, B.Sc., Ph.D. D. A. Govier, B.Sc., Ph.D.
Sugar Beet	V. McM. Davey. A. Murray (*resigned* 31/1/60). *Vacancy.*
Technical Assistants	A. McFarlane. W. Brown. A. P. Chatham, S.D.A. Mrs A. W. Chatham. Miss R. Cruickshank. A. Young. Miss M. D. Black (*appointed* 1/8/59).
Secretary	R. J. L. Gallie.
Assistant Secretary	Miss A. Malcolm.
Clerical Officer	Mrs H. H. Gray.
Clerical Assistant	Miss I. Rennie.
Shorthand Typist	Miss R. Jackson.

DIRECTOR'S REPORT

Brassica Crops.—The advent of a relatively uniform variety of thousand-headed kale known as " Canson " has drawn attention to the great variation in characters shown by some other commercial varieties of thousand-head. It has also become clear that the economic suitability of hybrid derivatives of kohl-rabi or broccoli crosses depends very largely on the choice of the kale parent. A new series of selections from thousand-headed kale crops was started in 1957, the chosen plants being kept alive by cuttings while their progenies were being examined in trials. Progenies of three kinds were obtained: self-fertilised by bud pollination, hybrids from controlled crosses between two known parents, and polycrosses where the seed borne on a mother plant had been pollinated by any of a group of selected kale plants. Seed of the last two categories was used for the trials. There were three trials, each of a different spacing: one where plants were set out at 24 in. × 24 in. intervals was used chiefly for the testing of controlled crosses in which relatively little seed was available; another contained mainly polycross samples, sown on ridges 26 in. apart, seedlings being singled to 10 in. intervals; and the third contained nine polycross progenies and a sample of commercial kale, all seeded in drills at the rate of one seed per inch.

Notes have been taken on side-shoot development, and, although it is too early to decide how this character is inherited, some of the progenies reflect the effect of the initial selection in favour of early branching; indeed there seems some risk of running to an extreme where the side branches are too long and woody. However, under the conditions of the third trial there was seldom any side-shoot development. In this trial all but one of the progenies were significantly shorter than the commercial control, which suggests a response to selection of short-stemmed parents rather than an effect of inbreeding, for there should be little inbreeding in these polycrosses. Although stem weights did not differ significantly there was a wide range of leaf weights, and the polycrosses of a selection TH-54, despite its shorter stem, yielded significantly more leafage than the control.

Crosses between thousand-headed kale and spring cabbage have reached the F_2 generation. So far there has been little segregation of attractive types; most were like loose cabbages,

though some hearted more strongly. Early bolting to flower is likely to be a nuisance in this kind of cross.

Recently a number of interspecific crosses were attempted involving various combinations of *B. oleracea*, *B. campestris* (including ssp. *chinensis*, ssp. *pekinensis* and ssp. *nipposinica*), *B. napus*, *B. juncea*, *B. carinata* and *B. hirta*. Many crosses were unsuccessful whilst others yielded only maternal plants. It had been suspected that stigmatic papillæ inhibit the germination of foreign pollen and the growth of pollen tubes, so in some crosses stigmas were removed and pollen applied direct to the cut surface of the style. This method did not increase the seed set, however. Suspected hybrids are being raised and their true identity ascertained through chromosome counts. One plant derived from the cross *B. napus* × *B. campestris* ssp. *nipposinica* was morphologically distinct from the rest of the progeny and proved to be a spontaneously doubled form with 58 chromosomes.

Autotetraploids of *B. oleracea* (mostly thousand-headed kale), *B. campestris* varieties and *B. hirta* have been produced by means of colchicine. Several techniques were attempted. Seeds soaked in various concentrations of colchicine either failed to germinate or produced seedlings with very severe root inhibition, mortality was high and in the survivors growth was extremely slow. The best results were obtained by spacing seedlings in boxes and applying colchicine solution dropwise to the stem apices at the cotyledon stage. Seedlings were treated in batches of 50, those showing distortion or inhibition were grown on, the rest discarded. Chromosome counts showed that for all concentrations 30-40 per cent of the plants retained were doubled. Seedlings which showed the most marked initial inhibition contained the largest proportion of tetraploids. A few plants were found to contain both diploid and tetraploid tissue whilst two individuals possessed some octoploid cells. The newly produced autotetraploid *B. hirta* ($2n = 48$) showed varying degrees of tetravalent formation at meiotic metaphase I. in P.M.C.'s. On the whole chromosome pairing was good and in some cells only bivalents were observed. Autotetraploid *B. campestris* ($2n = 40$) also showed fairly good pairing, the number of bivalents per cell ranging from 16 to 18. 4x *B. campestris* ssp. *chinensis*, ssp. *nipposinica* and ssp. *pekinensis* showed a similar range of pairing. Pollen fertility was fairly high (80 to 98 per cent) for all autotetraploids examined.

Progeny of the cross *B. napus* (2n = 38) × *B. campestris* ssp. *nipposinica* (2n = 20) were examined at mitosis and counts of 2n = 29 obtained. At meiotic metaphase I. ten bivalents and 9 unpaired chromosomes were found. Such pairing is to be expected in view of the fact that *B. napus* is an amphidiploid and already contains chromosomes derived from *B. campestris*. Hybrids between typical *B. campestris* and the oriental forms, ssp. *chinensis*, ssp. *nipposinica* and ssp. *pekinensis* (sometimes regarded as separate species), showed only bivalents at meiotic metaphase.

Various wild-types of *B. oleracea* and *B. campestris*, collected in the Mediterranean area and obtained from Svalöf, appeared to hold little promise as fodder plants. The *B. oleracea* forms were, on the whole, low growing and sparsely branched with poorly developed leaves. Other undesirable characters included long petioles to the leaves as well as a high stem to leaf ratio. *B. campestris* wild-types were all poor in growth and flowered early. Oriental forms of *B. campestris* were grown for observation. Most were of the early maturing type and produced little leafage. Two cultivars of *B. campestris* ssp. *nipposinica* (sometimes known as *B. japonica*) proved to be of interest. These developed rapidly and formed large rosettes with very many, rather finely dissected leaves. There was considerable variation in the time of flowering, from seed sown in early June most plants remained vegetative and retained their leaves for a long period. Other species in the *Brassicae* (*Eruca sativa, Crambe maritima, Diplotaxis muralis, D. tenuifolius* and *Raphanus rostratus*) have been grown. Apart from *R. rostratus*, a vigorous growing radish, all these species were poor in leafage and general growth. *R. rostratus* has been crossed with *B. oleracea* var. *fruiticosa* (thousand-headed kale) and the hybrids raised. So far *B. wrightii* (Lundy cabbage) and *B. monensis* (Isle of Man cabbage) have not been obtained.

Club-root.—The club-root fungus (*Plasmodiophora brassicae* Woron.) has long been the scourge of many brassica crops. Attempts have been made in the past to isolate resistant plants from field plots or boxes of infected soil. It has frequently been difficult to distinguish between actual resistance and escape from attack due to unevenness of infection. With this in mind, spores of *P. brassicae* have been isolated by grinding infected roots with water in a blender, passing the liquid through a filter and centrifuging the filtrate to obtain con-

centrated spore suspensions. Counts have been made, using a hæmocytometer, to determine the actual spore concentration of isolates. Once a standard technique of applying known quantities of spores to seedlings has been developed it is proposed to test all material, and particularly wild-types, in the hope of finding a source of immunity to the disease which would be useful in further breeding work.

An investigation into the uses and productivity of certain varieties of rape and kale is at present being conducted by the West of Scotland Agricultural College at Auchincruive, and in response to a request by the College for information regarding the botanical characters of the available rape types the Plant Breeding Station undertook to examine a number of varieties in a trial at Pentlandfield. The results of the tests appear in one of this year's Occasional Papers (page 45).

Herbage Plants and Genecology.—During the past three years the members of the section have devoted a considerable amount of time and thought to a genecological investigation of certain hill species collected mainly from the uplands of Southern Scotland. This investigation is now being analysed and it seems worth-while to put on record remarks and information which might be out of place in the final publication of results. There has been a growing distrust within the section of the generally accepted use of the within-population variance as the error item in the statistical analysis of genecological trials and this has been discussed in recent Occasional Papers published in the Reports for 1958, 1959 and elsewhere. A definite suggestion was made that the variance "between populations from ecologically similar sites" would provide a statistically more satisfactory error item, especially as a re-examination of some of our earlier results indicated that it might often be low enough to be useful. A new method of collecting had to be adopted which involved sampling a large number of sites, and this occupied the entire section very fully during a considerable part of one summer. Over two hundred sites were sampled largely in an area south-west of Edinburgh, and, as far as possible, eight plants of each of the following taxa were collected: *Agrostis* spp. *Festuca ovina*, *Potentilla erecta* and *Plantago lanceolata*. The small size of the sample is regrettable for several reasons, but there is a limit to the number of plants which can be grown and on which the

necessary observations and measurements can be made. However, for the purposes of this investigation the emphasis had to be on sampling a large number of sites. It should also be pointed out that the small area of some of the sites in itself imposed a limitation upon the number of tillers which could be collected since, in an attempt to avoid genotype reduplication, it was decided to sample at not less than ten-yard intervals, and many sites could not have yielded more than eight sampling points under these conditions.

The sites were classified into the following ecological types, and sites belonging to the same type were only sampled when at least four miles apart :—

A Species-rich bent fescue.
B Species-poor bent fescue.
C *Nardus*-dominated.
D *Molinia*-dominated.
E *Calluna*-dominated.
E′ Regenerating *Calluna*.

Most sites can be placed with confidence in one or another type, but those which cannot be so classified are being omitted from any analyses based on ecology. There is no pretence, of course, that all the *Calluna*-dominated sites exist under the same environmental conditions, but it is felt that they have something in common which distinguishes them from the other types even though that something must often remain undefined. It is obvious, though, that the species-rich bent fescue types occur on the more fertile soils, the *Molinia* on the much wetter ones.

The analysis adopted for each character scored is the simple analysis of variance between groups using the site means as the primary data. One attractive feature of this type of trial is that the same series of site means can be classified in several different ways, each one leading to its own analysis. Thus by grouping the sites by altitude (or geography, climate, &c.) an estimate can be obtained of the importance of this factor in population differentiation. Most of the work so far completed has adopted the ecological classification described above.

A major initial difficulty arose in connection with both the *Agrostis* and *Festuca* plants in that the collected material was not taxonomically uniform and much time had to be expended breaking down the complexes into their constituent parts. Both *A. tenuis* and *A. canina montana* were collected for two reasons.

In the first place, although typical specimens of either species are easily identified, it was impossible to be certain of those which appeared to have intermediate characters. Again, the two species tend to have definite ecological preferences and it would not have been possible to collect either *A. tenuis* or *A. canina* from the whole range of habitats. In the case of *Festuca* material it was known that the two chromosome races would most probably be collected but there was no means of distinguishing diploid from tetraploid in the field with certainty. The cytological examination showed that, in addition to the diploid and tetraploid races, a few triploids and other numbers were also present. A full account of this work was given last year and it was stressed that in both genera the proportions of the two constituent taxa change for the different ecological types and in both cases D is at one extreme and A at the other : D has the highest proportions of *A. canina montana* and the diploid fescue, whilst in A *A. tenuis* and the tetraploid fescue predominate.

A further difficulty in connection with this trial was the unfortunately high incidence of pests or diseases. The first to appear was rust on the *Agrostis* plots and before an adequate degree of protection could be found, it had caused considerable losses particularly among *A. canina montana* plants. Some plants of *P. lanceolata* showed signs of eelworm attack and they became more obvious in the second year when *Taphrina* also rather severely damaged some plants of *Potentilla*, though again a measure of control was eventually obtained, while *Festuca ovina* showed symptoms of barley yellow-dwarf virus. In spite of these handicaps much valuable data was nevertheless collected, and in some cases the differences between ecological groupings are highly significant.

The stems or leaves produced in the original environment were measured at the time of collection on at least two-thirds of the material, and these figures have been analysed for differences between site groups. The measurements did not give rise to any unexpected results since, for example, all plants collected from *Molinia*-dominated sites were tall and long-leaved, while, at the other extreme, plants from the species-rich bent fescue produced short leaves and very short, if any, stems. The analyses showed that the differences were statistically significant, and it is interesting to record that there is a definite tendency within a species for the wild measurements

to be closely correlated with measurements obtained from the same populations in cultivation, although naturally the differences between the tall and short, for example, are much less in the latter case. Furthermore, there are correlations in the cultivated material between species; for example, the leaf length of tetraploid fescues is positively correlated with the leaf length of plantain and also the plant diameter of *Potentilla*. Sites with genetically big plants of one species tend to have genetically big plants, in one sense or another, of the other species, and so it seems clear that the several species are acting in a parallel fashion to the same or very similar environmental influences. These highly significant correlations can be determined without imposing any kind of classification on the sites, but it is interesting to note that the correlations within ecological types are very considerably reduced, indicating that the classification adopted was efficient at breaking up the range of the important environmental factors into reasonably homogeneous units.

Work on the natural spread of clones of *Trifolium repens* has yielded some interesting results. It appears that in this species clones are sometimes distributed discontinuously on the ground. Furthermore, the different patches of one clone seem to have some ecological parity, the floristically distinct intermediate zones frequently containing other genotypes of clover. However, in the absence of any known mechanism of vegetative spread that could cover several yards in one step, it seems obvious that the intermediate zones must have been occupied at some time. The indication of marked ecological segregation of the various clones suggests that they are very specialised in their requirements, and this contrasts with our findings on grass genotypes.

In connection with the investigation of lead tolerance in *Festuca ovina* many progenies produced by crossing plants of differing tolerance have now been tested, and it is clear that a simple major gene with two alleles would be insufficient to explain all the results. As suspected earlier high tolerance is often completely dominant over non-tolerance, but in some crosses this dominance did not appear, and the various types of result which have been recognised so far are discussed in more detail in an Occasional Paper in this Report.

In order to improve the aeration of the roots and the mixing of the solutions when testing for lead tolerance a hoisting

mechanism has been installed in the controlled environment chamber which raises the tubes out of the liquid for 40 minutes in every 80. The atmosphere in the tubes remains sufficiently damp for the roots to continue growing freely. The first tests of this mechanism suggest that it might lead to a substantial fall in the error variance of the tolerance measurements.

At the same time the search has continued for a completely new method of measuring tolerance, and preliminary experiments were conducted on the culturing of excised root tips, the counting of cells from macerated roots, and the effect of lead on the invertase activity of the root tip. It was found difficult to obtain sufficiently sterile conditions for the culture of root tips taken from growing tillers. Many tips grew well for several days, but others became seriously contaminated, and although streptomycin successfully controlled bacterial growth, no fungicide of comparable value was found. Root cells were counted, after maceration in chromic acid, by using a hæmacytometer chamber with dark-ground illumination. The results suggested that lead reduced both the number and the size of cells, as expected, but no clear differences were found between tolerant and non-tolerant plants, and in any case it was felt that the labour involved would make the method unsuitable for the routine examination of large numbers of plants.

The measurement of invertase activity gave more promising results. A colorimetric method of determining glucose with alkaline sodium picrate enabled that produced by single root tips in sucrose solution to be estimated after a few hours. The invertase activity was found to be reduced (*a*) by the pre-treatment of the intact root with lead nitrate, and (*b*) by the presence of lead in the sucrose solution itself. In both cases the activity in a tolerant plant was reduced less than that in a non-tolerant plant, but the difference was particularly marked with the pre-treatment method. The possibility of using this as a routine test for tolerance is being investigated.

During the past year the effect of ryegrass mosaic virus on the yield of Italian ryegrass was estimated in a field trial, and the fescue virus already mentioned as causing damage to plants in the genecological trial was successfully transmitted to healthy plants of *Festuca ovina*. Further reference to the grass virus investigations appears on page 18.

Oats.—The practice of making selections from unfixed hybrid material in three distinct environments at centres in Argyll, Inverness-shire and Pentlandfield has been continued. The value of this procedure has been borne out by the results of a series of trials conducted at the three centres in order to assess the relative performance of locally selected lines.

The trial at the Argyll centre contained forty-two fixed lines of which twenty-one had originated in Argyll, six in Inverness-shire and the remaining fifteen at Pentlandfield. In preliminary trials at Pentlandfield in 1958 twenty-two of these lines had equalled or bettered the control in grain yield, but only one of these had been selected at the Argyll centre. In 1959, however, at the Argyll centre, the controls were exceeded by thirteen of the twenty-one Argyll lines, which included the two highest grain yields in this trial, three of the six Inverness lines and eight of the fifteen Pentlandfield lines.

At the Inverness-shire centre forty-two lines were tested, of which seventeen had originated in Inverness-shire, four in Argyll and twenty-one at Pentlandfield. In 1959 the controls were exceeded by eleven of the seventeen Inverness-shire lines, again including the highest grain yield in the trial, by all four Argyll lines, and by seventeen of the twenty-one Pentlandfield lines.

At Pentlandfield, the trial contained sixty lines and included all those tested at the other two centres. In this trial only six lines gave a higher grain yield than the control, three of these being from Pentlandfield, two from Inverness-shire and one from Argyll.

From the foregoing results it is evident that selections made in the difficult Argyll and Inverness-shire environments were relatively more successful at their respective centres of origin than the other lines which had been selected under environments prevailing elsewhere. Moreover, the lines which at the Argyll and Inverness-shire centres gave the highest grain yield were also those which produced the greatest bulk of straw, suggesting that the short-strawed, high grain-yielding types which are successful in the Pentlandfield environment are not the best agronomic proposition for the Argyll and Inverness environments.

During the year upwards of 40,000 seedlings have been tested for resistance to oat-stem eelworm, from which about 1,000 highly resistant ones have been retained for multiplication and

field trial. In the course of this work it was observed that the symptoms obtained in susceptible and partially resistant material were much more severe than hitherto observed. Since the eelworm used for inoculation purposes in each year were freshly collected from infected crops from as far apart as Edinburgh and Orkney, it was thought likely that the observed differences in symptoms were due to the occurrence of different eelworm races.

In order to determine whether in fact this was the case the cultivars Early Miller, Milford and Albyn Bard, representing the tolerant, resistant and susceptible reactions respectively, were inoculated at the rate of 300 eelworm per seedling with four different lots of eelworm which were (*a*) collected near Stonehaven in 1958 and used in the 1958-59 experiments, (*b*) the same, after multiplication for a year in fodder beet, (*c*) collected at Fordoun near Laurencekirk in 1959 and (*d*) collected at Burdiehouse near Edinburgh in 1959 and used throughout the inoculation experiments in 1959-60. Twenty-one days after inoculation there were marked differences in the severity of the symptoms exhibited by the inoculated plants according to the eelworm used. In each cultivar the order of severity was the same, the least affected being the seedlings inoculated with (*b*) which only differed slightly from (*a*) while those with (*d*) were the most affected. Intermediate between these two levels were the seedlings inoculated with (*c*). What is noteworthy is that the selections previously regarded as resistant have stood up to the particularly virulent Burdiehouse race.

A search has also been made for new sources of resistance. A high degree of resistance has been found in the following :—

Station Reference No.	*Name*	*Source*	
Aa 94	Algerian (Red)		
Aa D. 5	*Avena sterilis*	Spain	
Aa D. 7	,, ,,	,,	
Aa E. 7	,, *byzantina*	Macedonia	Greece
Aa E.12	,, ,,	Eria Eubosa	
Aa E.13	,, ,,	Peloponessus	
Aa E.18	,, ,,	Epiros	
Aa E.19	,, ,,	Peloponessus	
Aa E.35	Yezilkog No. 138 (*Avena byzantina*)	Turkey	

It will be noted that all the above fall into the same category as *Avena ludoviciana*, the source of resistance in the hybrids

at present under test, the new resistant types being either the wild *Avena sterilis* or cultivars derived from it.

Potatoes.—The Merit Trials conducted by the Department of Agriculture for Scotland in 1959 contained 13 of the Society's seedlings, three in the 2nd Year and ten in the 1st Year Trials. Of these one seedling was recommended for further trial in the 3rd Year, and six in the 2nd Year in 1960. For the 1st Year Trials in 1960, ten seedlings have been entered. Stocks of all these seedlings were multiplied at Blythbank and tested for virus content during the growing season. No viruses were detected.

Stocks of Pentland Beauty and Pentland Crown, grown at Windyedge, Perth and Blythbank, Peeblesshire, respectively, were graded "V.T.S. (Scot.)" by the Department of Agriculture for Scotland. These stocks have now been transferred to the agents for further multiplication and marketing. A further stock of Pentland Crown of approximately two acres which was grown in Perthshire was graded "A (Scot)."

In the past it has been the custom to send samples of seedlings abroad for trial, particularly under tropical conditions where blight can be a serious problem. Some of these seedlings have given promising results in Kenya and are now grown on a commercial scale. A request has been received from the Department of Agriculture in Kenya to name five of them and this matter is receiving attention. One of these seedlings, Ref. No. 1565 (4), has also given promising results under commercial conditions in Cornwall, following trials by the National Agricultural Advisory Service, and it has been entered for the Wart Disease and Identity Test of the Department of Agriculture for Scotland with a view to registration.

The investigations on breeding for field resistance to blight were continued during 1959. About 270 seedlings which survived the screening tests with race (1, 2, 3, 4) in previous years were grown in field plots to assess their economic potentialities and to expose them to the disease under natural conditions. In the unusually dry season, however, no blight was observed in the plots. Samples of most of these seedlings were sent to Dr J. S. Niederhauser, The Rockefeller Foundation, for test in the Toluca Valley, Mexico. The results obtained by Dr Niederhauser showed that while the resistance to race

(1, 2, 3, 4) observed at Pentlandfield remained fully effective in some of the seedlings under Mexican conditions, it did not do so in others. It is now clear that those reacting with susceptibility in Mexico had been protected against race (1, 2, 3, 4) at Pentlandfield by at least one additional R gene (*e.g.*, R_5) inducing hypersensitivity. With the help of these results, the true " field resisters " have been distinguished and valuable information on the lines to follow in further breeding experiments has been obtained. The most promising of this material originated from interspecific hybrids involving *S. demissum*, *S. phureja* and *S. simplicifolium*. Further progenies comprising some 4,000 plants were screened with race (1, 2, 3, 4) in 1959 and the survivors retained for similar treatment.

On account of the relative scarcity of blight in the field plots in 1959 few samples were taken for identification and only races (3), (4) and (3, 4) were recorded.

Samples of blight from overseas identified in 1959 numbered 269. The majority of them were received in connection with surveys of the distribution of blight races occurring in Italy and Brazil in co-operation with Professor Antonio Ciccarone of the University of Bari and Dr B. Bastoz Cruz, Instituto Biologico, São Paulo. In Italy race (0) was found to be prevalent on tomatoes and race (4) on potatoes. Races (3), (2, 4) (3, 4) and (2, 3, 4) were also identified but were relatively uncommon.

In Brazil, potatoes were affected mainly by races (4) and (1, 4) while tomatoes were attacked by races (0), (3) and (3, 4). Races (1, 3, 4) and (2, 3, 4) also occurred but were very limited in distribution.

In the course of raising new seedlings at Pentlandfield in 1959 two unusual diseases were observed in the greenhouse. One was a mildew caused by forma *matricalis solani tuberosi* of *Erysiphe polyphaga* Hammerlund. The other, a bacterial disease caused by *Bs. subtilis*, was evident in some progenies but not in others. An Occasional Paper dealing with this disease appears on page 24.

In evaluating resistance to potato root eelworm for breeding purposes it is necessary to decide upon a " standard " level of resistance to aim at in new varieties of potato and to classify the resistance of particular clones in relation to this standard. Such a classification can also serve as a basis for tabulating strain relationships. The classification adopted at Pentland-

field and the status of physiological strains of potato-root eelworm are discussed in an abstract on page 22.

The relationship between the resistance in twelve species of potato to the Duddingston and Boghall strains of the eelworm is summed up in the following Table :—

Series	*Species*	*Strain reaction*	
		Duddingston	*Boghall*
Tuberosa	*S. tuberosum* subsp. *andigena*	+	−
	S. famatinae	+	−
	S. macolae	+	−
	S. microdontum	+	−
	S. gourlayi	±	−
	S. vernei	−	−
Cuneolata	*S. infundibuliforme*	−	?
Megistacroloba	*S. sanctae-rosae*	−	−
	S. raphanifolium	±	+
	S. megistacrolobum	±	+
Tuberosa	*S. neohawkesii*	±	+
	S. multidissectum	−	+

Level of resistance	*Sign*	*Strain reaction*
above standard	−	non-aggressive
near standard	±	indefinite
below standard	+	aggressive

The resistance in the species tested above is characterised on the basis of the few clones of each species which have been tested for resistance to date. Clearly, a high level of resistance to eelworm is not an uncommon feature of the many species of potato indigenous to South America.

Survivors of the collection of tuber-bearing *Solanum* species received from Dr Hawkes in 1958 were tested for the presence of viruses. Strains of virus Y were detected in 14 units and viruses X and S, alone or in combination, were found in 15 units. The remaining 123 units appeared to be virus-free and the collection will now be examined for characters of potential value in potato breeding.

In breeding for resistance to viruses, over 4,000 seedlings were inoculated with X and Y viruses in the glasshouse and the susceptible seedlings discarded. Over 2,000 seedlings remained and were planted out in the field. At harvest, losses and elimination of seedlings with undesirable characters reduced this material to 261 plants for further trial.

The natural spread of aphis-borne viruses at Pentlandfield

was too low in the years 1955-57 to permit reliable assessment of resistance to leaf-roll and virus Y under field conditions. In 1958, therefore, trials were conducted at Cambridge and Musselburgh as well as at Pentlandfield. There were 480 plants, comprising 12 varieties, in each trial and the spread of infection was evaluated at Pentlandfield in 1959.

There were overall infections of 6 per cent Y and 5 per cent leaf-roll at Pentlandfield, 12 per cent Y and 69 per cent leaf-roll at Cambridge and 12 per cent Y and 89 per cent leaf-roll at Musselburgh. At Cambridge, differences in varietal susceptibility to leaf-roll were reflected in the range of infection from 27 per cent in Pentland Crown to 83 per cent in the susceptible variety Majestic, but at Musselburgh these differences were largely suppressed by the overall high rate of infection and at Pentlandfield there were too few infections to make an assessment. The spread of virus Y was too low at all centres for the adequate evaluation of varietal differences towards this virus.

Pentland Crown had fewest infections with both leaf-roll and virus Y at each centre and it would seem, therefore, to possess resistance to both viruses. None of the other seedlings in the trials was recommended for naming.

Forty-four lines of *Solanum demissum* were examined for their reactions to virus Y and virus A. Three lines were found to be homozygous for the gene controlling necrotic response to both viruses, 29 lines were homozygous for necrotic response to virus A only and 8 lines were non-necrotic and susceptible to both viruses. The remaining 4 lines were heterozygous.

In preparation for further studies on strains of virus Y, 22 cultures were tested on differential hosts. Two cultures were found to contain a mixture of strains whilst the other remained true to type.

In addition to the study of potato viruses the activities of the Section have recently been extended to include a preliminary examination of the virus situation in herbage grasses. As part of this programme the effect of ryegrass mosaic on the yield of S22 Italian ryegrass was estimated in a field trial which contained 40 healthy plants and 40 diseased plants of each of two clones. The material was cloned, and part of each clone inoculated, in the summer of 1958. The plants were kept in the glasshouse over the winter, further multiplied in the spring

and the trial was planted on 15th May 1959. Three harvests were made during the season.

In the harvests made on 16th July and 21st August, the diseased plants of clone 1 yielded respectively 56.5 per cent and 59.5 per cent of the healthy plants and the diseased plants of clone 2 yielded 18.4 per cent of the healthy plants on each occasion.

On 21st August it was noticed that infection had spread to a few of the healthy plants of clone 1. The " healthy " plants of both clones were carefully examined on 29th September and all those of clone 1 were at least partly infected. Plants of clone 2 had shown only a mild mottle following inoculation the previous year and the severe stunting responsible for the large yield reduction had developed after the overwintering period. Similarly, when examined for cross-infections, some of the " healthy " plants of clone 2 bore mottled tillers but the symptoms were too mild to make a reliable assessment of the number of plants infected. Ryegrass mosaic virus is transmitted by mites and, allowing about three weeks for symptoms to appear, most of the spread must have occurred during August. Only a few mites were found on the plants at the end of September.

When the third harvest was made on 14th October, the yield of the diseased plants of clone 2 was 21.3 per cent of the healthy plants, a figure in agreement with those for the previous harvests. The diseased plants of clone 1 yielded 85.6 per cent of the healthy plants; that this high figure was entirely attributable to the effect of cross-infections in the " healthy " plants was apparent from the absolute yields, the yield of the diseased plants being comparable with their yield in the earlier harvests whereas the yield of the " healthy " plants was considerably less.

The plants in the trial were spaced 3 ft. apart and it is probable that the reduction in yield would be much less pronounced in a sward where infection would rarely reach 100 per cent and where healthy plants could fill the spaces left by adjacent, less vigorous, diseased plants. Further trials are being made with a wider range of ryegrass material.

An antiserum to ryegrass mosaic virus proved to be of limited value because it reacted only with material from young vigorous plants showing well-marked mottle symptoms.

Work on the aphid-transmitted viruses of cereals and grasses

was hampered by virus-contamination of the aphis culture, but the fescue virus reported last year was transmitted to healthy plants of *F. ovina* and, when these were planted in the field, they showed the stunting and reddening of the leaf tips that characterised the plants in which the virus was originally found. No oat plants showing definite symptoms of barley-yellow dwarf were seen in any of the oat crops examined.

Sugar Beet.—When the Station undertook work with sugar beet most attention was given to the material already bred by the Cambridge Plant Breeding Institute, which was then being tested for yielding capacity as well as resistance to bolting. However, in response to a request for a variety with large tops, two groups of selections were taken from commercial crops to form the basis of a breeding programme. The first selections were made in 1948 and involved 630 plants from an early-sown crop at Logie Farm, Newburgh, Fife. These were then reduced to 50 by tests of specific gravity and root-weight. A second group of 100 plants was chosen in 1949 from a crop at Scotscraig Mains, Tayport. These two groups of plants were seeded, and the progenies of mother-plants were compared. Lines or "families" were bred from the best of these by seeding groups of selected plants in separate isolation plots.

It has been an accepted practice of Continental breeders to form a strain by mixing the best families at the end of ten years' breeding and testing. The Sugar Beet Research and Education Committee requested the Station to do this with what remained of the Logie families. In 1958, therefore, seed-beds were sown to provide stecklings of all the families which might ultimately be included in an experimental strain, and trials were laid out to test all the families for which seed of good germination vigour was available; both inbred lines and polycross groups being tested. As a result eight families were chosen for inclusion in an experimental strain, and stecklings of these were sent to Cambridge, where they were intermixed in a large isolation plot, and over 1 cwt. of seed has been obtained. The two best families were, and still are, Logie-5 and Logie-F, and seed from a mixture of stecklings of these was also harvested at Cambridge in 1958. Another mixture seeded there contained both Logie families and three families

from a Cambridge strain. Sufficient seed of the latter mixture was available for inclusion in some small trials arranged by the National Institute of Agricultural Botany in 1959. The Logie material has also been used in attempts to obtain families with monogerm seed and for crossing with male-sterile sugar beet.

In 1954 the Station was asked to make a new attempt at combining large-foliage, good yielding and high-resistance to bolting. Since intra-varietal selection had proved ineffectual it was decided to intercross varieties of large shaw type, and to attempt to select from among the hybrids. Pairs of plants were isolated in glasshouse compartments and gave small quantities of seed which were sown out in replicated strips in 1956. Following specific gravity tests selections were made of the plants from the four best units. Four plants of each unit were seeded together in a glasshouse compartment, and gave sufficient seed in 1958 to lay out a small yield trial. After a mid-April sowing no bolters occurred. Three of the lines had foliage of almost identical type, with leaves which were dark-green, short-stemmed and too small. The fourth line had a large, light-green foliage of very distinctive appearance, but when the trial was lifted its roots were found to be very small and highly fanged. The object of combining yield of root and top had not been achieved.

A further attempt was made to obtain a large-foliage variety and this involved selections from each of four supposedly large top varieties, Bush E, British S.K.W., Cesana and Italian 8. About ten plants of each variety were inter-crossed in a glasshouse. As a result of the progeny tests conducted in 1956 four groups were chosen, two from Bush, one from British S.K.W. and one from Cesana. These were seeded together in an isolation plot, and the bulked progeny was compared with commercial varieties in a field trial. The result was again disappointing, for, while the sugar percentage equalled that of the controls, the weight of tops was insignificantly lower and the root weight significantly so.

Several samples of other lines, derived from experiments laid down in 1954, were compared in trials conducted in 1958 at Gosford, East Lothian, and at Pentlandfield. One family, ENBL-18, gave very promising results for yield and resistance to bolting, but at both centres the top weights were hardly up to the trial averages.

Publications

DUNNETT, J. M. (1960). Potato breeders' strains of potato root eelworm (*Heterodera rostochiensis* Woll.). *Nematologica*. (*In press.*)

A selective host for an aggressive population of potato root eelworm may be defined as a clone or variety of potato possessing resistance which discriminates against the survival of other populations of the parasite. There will be a trend towards homozygous aggressiveness in a population multiplied repeatedly on a selective host. Such a population may be regarded as a breeders' strain. Different strains will have different selective hosts, together comprising a differential host series. It is proposed to restrict the differential host series to plants possessing at least a " standard " level of resistance to one or more strains. Standard level of resistance is specified as that resulting in 90 per cent of the plants of a clone remaining cyst-free on the basis of soil-ball inspection after exposure to infection in 4-inch pots of culture medium of 40 eggs/gm. infectivity. The proposed categories of resistance are (1) " above standard " (— sign) constituting the level of resistance aimed at in new varieties of potato; (2) " near standard " (± sign) covering the range between clones with 90 per cent of the plants cyst-free and clones with a mean cyst-count of 10 cysts per soil-ball; (3) " below standard " covering the remainder of the range to maximum susceptibility, corresponding to a cyst-count of 1000-1250 cysts per soil-ball. In tabulating strain relationships the — sign means " resistant " (with reference to the plant) or " non-aggressive " (with reference to the parasite) and the + sign means the corresponding " susceptible " or " aggressive." The ± sign indicates an indefinite strain reaction. Plants possessing the resistance of *Solanum tuberosum* subsp. *andigena* C.P.C. 1673 and *S. multidissectum* are selective hosts for the aggressive Duddingston and Boghall strains respectively. Neither strain contains more than a negligible proportion of biotypes which develop to female maturity in the selective host of the other.

HARBERD, D. J. (1960). Association-Analysis in Plant Communities. *Nature*, **185**, 53-54.

In subdividing a population of species lists, the advisability of looking for and relying on that single species which does the job most efficiently by its presence or absence is questioned. A technique which has been found useful at this station is introduced; the distance, called d^2 between two site lists, is taken as the number of species represented once only in the two lists. Grouping a complete table of d^2 to give low mean values within groups relative to that between groups arranges the floristic information in a meaningful manner.

WILKINS, D. A. Recognising Adaptive Variants. *Proc. Linn. Soc. Lond.* **171**. (*In press.*)

Summary.—

The real adaptive value of big morphological differences between taxa cannot be studied experimentally. The smaller differences found within a breeding unit, the most important of which may be physiological, are open both to genetic analysis and to experiments for comparing the fitness of two types in a chosen environment. The most direct experiments entail competition between the types in a natural habitat, and a controlled environment can only be substituted if the subsequent extrapolation is unambiguous. The indirect method of finding genetic differences by uniform cultivation and correlating these with differences between wild habitat factors is valuable, but is subject to some uncertainty about the choice of characters and factors to measure.

Lead tolerance in *Festuca ovina* was studied as an example of intense selection without direct competition. Soil contaminated with lead was poisonous to most plants of the species, but allowed a few to grow, and parallel results were obtained with root growth in culture solutions containing lead. The measurement of tolerance was too inaccurate for exact genetic analysis, but there was evidence of a major gene separating the plants into three types, and tolerance was dominant over non-tolerance. Similar degrees of tolerance were found in the two chromosome races and in plants from different isolated mining areas of Britain, so that it may have arisen more than once and may not always be due to the same gene.

In a garden trial no morphological differences between tolerant and non-tolerant plants could be found. It is suggested that the term "ecotype" should not be applied to a variant due to a single gene, but should be reserved for cases where a number of correlated characters separate clearly distinct races.

A DISEASE OF POTATOES AND TOMATOES CAUSED BY *BACILLUS SUBTILIS*

JEAN F. MALCOLMSON

In the spring of 1959 unusual symptoms were observed among potato plants in a greenhouse at Pentlandfield. Areas of the leaves were darkened, as though splashed with oil, and on the underside the veins of the affected parts were black and surrounded by watersoaked, dark green tissue. Within a few days the discoloured tissues became dried and brown, and severely affected leaves often wilted and dropped from the stems. When the leaf symptoms were apparent the axillary buds and shoots were dead and usually a die-back of the aerial parts developed. On tubers, particularly at the rose end the "eyes" were dead and occasionally the tissue adjacent to them was rotted.

Smears from the diseased tissue, and from apparently healthy tissue in the region of the vascular bundles of the stems and tubers, revealed the presence of Gram-positive, rod-shaped bacteria. On isolation these bacteria were identified as *Bacillus subtilis*, according to the scheme of Gibson and Topping (1938). The absence of bacteria other than *Bs. subtilis* in the isolation plates was particularly marked.

Isolates of *Bs. subtilis* obtained at Pentlandfield were tested for pathogenicity to potatoes using two methods of inoculation.

1. The stem was pricked at the base of a petiole with a needle which had been dipped in the inoculum; a needle dipped in sterile water was used to prick the controls.

2. With the aid of a sharp scalpel a narrow strip of epidermis between the "wings" of the stem was torn downwards for about 2 cm. A drop of inoculum was placed on the exposed tissue then the epidermis was replaced. Distilled water was used in place of the inoculum in the controls.

The first method was designed to introduce the bacteria directly into the vascular tissue while the aim of the second method was to avoid this.

Inoculum was prepared from four isolates, grown for 48 hours on nutrient agar, by suspending them in distilled water. Four plants per isolate and one plant per isolate were inoculated

by the first and second methods respectively. In each case an equal number of control plants was used and all the available stems were treated. The plants were of the variety Pentland Crown and they were about 30 cm. high when inoculated. Following inoculation they were kept in an unheated greenhouse under normal conditions.

After 10 days all but two of the plants inoculated by method 1 and two plants inoculated by method 2 showed typical signs of infection. By the end of 3 weeks all the inoculated stems were infected. One stem on each of two control plants was also affected but the symptoms appeared some 7 days before they were apparent on the plants inoculated with *Bs. subtilis*, suggesting that the bacteria had been in them before the experiment commenced. Otherwise the control plants remained healthy.

Bacteria identical with those used as inoculum were isolated from the region of the vascular tissue of the stems and tubers of the affected plants. Therefore, it was concluded that *Bs. subtilis* had caused the disease.

To determine whether diseased plants might arise from planting infected tubers twenty tubers from naturally infected plants were planted in pots and kept in a cool greenhouse. In ten of the tubers the " eyes " were normal and there was no sign of infection, while the remaining ten each had at least one dead " eye." All the plants from apparently uninfected tubers emerged and four appeared free from the disease after 3 months. Three produced stems from 7 to 30 cm. high but those were severely affected by the disease, and two emerged but collapsed before the shoots were more than 5 cm. high. Among the plants from obviously infected tubers, three produced stems greater than 15 cm. high but they were severely infected; four emerged but were severely infected and made no appreciable growth and three failed to emerge. Five tubers from healthy plants were used as controls and they gave rise to normal plants with no sign of infection within 3 months.

Since *Bs. subtilis* was isolated from potato tubers, and planting infected tubers gave rise to infected plants, it was concluded that the disease is tuber-borne.

In the late stages of infection the symptoms on plants infected with *Bs. subtilis* are similar to those of ring-rot, a serious disease of potatoes also caused by a Gram-positive bacterium, *Corynebacterium sepidonicum*. Since accurate

diagnosis of ring-rot by isolating the pathogen and reproducing the disease requires considerable time, other means of determination have been employed. Racicot, Savile and Conners (1938) suggested the preparation of smears from diseased stems and tubers, staining them by Gram's method and examining them microscopically for the presence of Gram-positive bacteria. Iverson and Kelly (1940) used ultraviolet light for diagnosing the disease, infection being indicated by fluorescence of the affected tissues in the tubers. Combination of these two tests has been recommended by Glick *et al.* (1944) but O'Keefe and Werner (1959) in selecting ring-rot free clones have found the ultraviolet technique alone to be as efficient as when it is used in conjunction with the Gram-staining method.

In smears prepared from the vascular tissue of tubers and stems of potatoes infected with *Bs. subtilis*, the Gram-staining method demonstrates the presence of minute, Gram-positive, rod-shaped bacteria. Such vascular tissue, when viewed under ultraviolet light shows a marked white to green fluorescence. Therefore, to distinguish the disease from ring-rot, *Bs. subtilis* must be obtained and identified in culture.

Bs. subtilis is more readily isolated in culture than *C. sepidonicum* and it can be clearly distinguished from the latter since it produces spores on culture media. However, the bacteria obtained from the host closely resemble *C. sepidonicum* as they are much smaller ($0.5 \times 0.9\mu$) and more variable in shape than those on culture media ($0.5 \times 1.5\mu$).

The pathogenicity of *Bs. subtilis* towards tomato plants was also investigated. Using method 1, two plants per isolate were inoculated then kept under normal conditions in a greenhouse. Two plants were treated as controls. In 6 days two or three leaves above the point of inoculation showed watersoaked tissue surrounding the marginal veins. After 7 days these veins became black and this darkening progressed towards the petiole while the watersoaked areas coalesced and dried out. Within 14 days the leaves shrivelled and usually dropped from the stem. The young leaves at the growing tips of the plants at first showed a marked chlorosis along the veins, but symptoms like those on the older leaves developed as the disease progressed. Brown streaks appeared on the stems within 18 days and in time they extended to 8 to 10 cm. in length. The two control plants remained healthy throughout the test.

Spore-forming bacteria are not usually regarded as plant pathogens but occasionally species of *Bacillus* have been reported to cause disease in plants. Rudakova, Staruigina and Shishelova (1950) recorded *Bs. subtilis* as the cause of a soft-rot of cabbage while Volcani and Wahl (1954) and Volcani (1956) attributed soft-rots of tomato, chilli and eggplant fruits to infection with the same organism. In each case a relatively high temperature (35°C., 33°C. and 27°-37°C. for cabbage, tomato and eggplant respectively) was considered necessary for the infection to develop.

In the previous records *Bs. subtilis* has been associated with a soft-rot of the affected organs but the infection of potato and tomato considered in the present paper was not associated with such symptoms. In addition, the temperature in the greenhouse in which infection occurred never exceeded 20°C. —a much lower temperature than was considered necessary for infection in the above records. Such a temperature could be expected to occur in the field in a normal season ; therefore, although the disease has so far been observed only in the greenhouse, temperature would not likely prove a limiting factor for its development in the field.

It seems that all potatoes are not susceptible to *Bs. subtilis*. Inoculation of Pentland Crown plants, for example, resulted in 100 per cent infection while Craigs Royal plants inoculated in the same way and with the same inoculum remained completely free from the disease. In addition, when the disease first appeared it was conspicuous among the progenies of Pentland Crown and Saco crossed with a seedling variety 2288a(2). A second crop in the same greenhouse was also infected, and again infection was limited to the progenies of seedling variety 2288a(2) crossed with four other seedling varieties and Saco. In both instances the disease was not restricted to any one area of the greenhouse and neighbouring plants, the progenies of different crosses, remained free from infection.

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RACES OF *PHYTOPHTHORA INFESTANS* AND RESISTANCE PROBLEMS IN POTATOES

WILLIAM BLACK

The severe epidemics of potato blight in Britain in 1845 and 1846 marked the beginning of a search for resistant varieties which has continued with a fluctuating measure of success until the present day. Genes conferring resistance to blight were not to be found in the original introductions into Europe from South America and it was not until 1909 that hereditary resistance was demonstrated in a wild species of potato (Salaman, 1912). About the same time, Dr Wilson of St Andrews introduced *Solanum demissum* into his breeding programme. Some of the seedlings he produced remained free from the disease under ordinary cultural conditions for a number of years but in 1932 blight appeared on these and other hybrid seedlings that had hitherto been regarded as immune. This apparent breakdown of resistance was found to be caused by a new physiological race of the parasite. A study of these plants revealed that the freedom from blight previously exhibited by them was not due to immunity but to the hypersensitive nature of the cells. The parasite penetrated the tissue but was sealed off at the points of infection by the rapid formation of a necrotic barrier. This hypersensitive response proved specific to particular races of the parasite. By repeated backcrossing to commercial varieties for the purposes of improving yield and other agronomic qualities, the genes controlling hypersensitivity were separated and later classified. The genes were then recombined as required by intercrossing appropriate genotypes of known constitution.

By this means a differential host series of sixteen genotypes based on four major genes R_1, R_2, R_3 and R_4 was gradually built up at the Scottish Plant Breeding Station. Progress was slow at first when few races of the parasite were available but later it was speeded up as isolates from overseas were received for identification. Among those of special interest at the time were races (1, 3) and (2, 4) from Dr G. B. Wallace, Tanganyika; (2) and (1, 2) from Dr R. M. Nattrass, Kenya; (3) and (3, 4) from Dr Bazan de Segura, Peru; (1, 2, 4) and (2, 3, 4) from Dr Mastenbroek, Holland; and (1, 2, 3) and (1, 2, 3, 4) from

Dr J. L. Howatt, Canada. With the help of these isolates a differential host series of sixteen plants capable of distinguishing sixteen different races of the parasite was completed. This scheme was adopted as the international system of classification (Black *et al.*, 1954). It soon became apparent, however, that the species *S. demissum* must possess more than four such genes, since certain clones and hybrid seedlings bred from them, proved to be field immune from race (1, 2, 3, 4) (Black and Gallegley, 1957). Field immunity from race (1, 2, 3, 4) was also observed in *S. demissum* and *S. stoloniferum* by Professor Schick, Gross-Lüsewitz, who kindly supplied samples of his host genotypes. Among them was a clone of *S. stoloniferum* labelled R_6, which proved to be hypersensitive to race (1, 2, 3, 4) but susceptible to the two races of blight from Holland previously classified as " 1, 2, 4 " and " 2, 3, 4." With the inclusion of the *S. stoloniferum* R_8 genotype these races were reclassified (1, 2, 4, 6) and (2, 3, 4, 6) respectively.

Race (1, 2, 4, 6) was employed to test a collection of seedlings bred from *S. demissum* while parallel tests were made with the Canadian isolate of race (1, 2, 3, 4). On the basis of the reactions obtained, the seedlings consisted of four different types :—

I. Hypersensitive to both races.
II. Hypersensitive to race (1, 2, 3, 4) but susceptible to race (1, 2, 4, 6).
III. Susceptible to race (1, 2, 3, 4) but hypersensitive to race (1, 2, 4, 6).
IV. Susceptible to both races.

In this experiment the essential genes were R_3 and R_6. The former induced a hypersensitive response to race (1, 2, 4, 6) and the latter a hypersensitive response to race (1, 2, 3, 4). These results are summarised in Table 1.

The evidence shows that *S. demissum* possesses a gene comparable with the R_6 gene recognised in *S. stoloniferum* and that two races of the fungus pathogenic to the R_6 genotype have been in existence for several years. A further race, (2, 4, 6) was identified in 1959. It appeared as a mutant in a culture of race (1,2,4,6) which had been maintained throughout the winter in tubers of the variety Kerr's Pink. Apparently this isolate had lost the power to attack the R_1 genotype during the period of culture in *S. tuberosum* tissue.

TABLE 1

REACTION OF *S. DEMISSUM* DERIVATIVES TO RACES (1, 2, 3, 4) AND (1, 2, 4, 6)

Type	Race (1, 2, 3, 4)	Race (1, 2, 4, 6)	Essential genes
I	–	–	R_3R_6
II	–	+	R_6
III	+	–	R_3
IV	+	+	r

– = hypersensitive + = susceptible

Recent reports from Canada (Eide *et al.*, 1959) indicate that the differentiation of blight races has gone a step further. Three new races designated (1, 2, 3, 4, 5); (1, 2, 3, 4, 6) and (1, 2, 3, 4, 5, 6) have been isolated from juvenile or senescent leaves inoculated with race (1, 2, 3, 4). The differential host series should accordingly be extended to include six genes which in their different combinations will constitute a series of sixty-four different genotypes and will be capable of distinguishing sixty-four different races. Since *S. demissum* is hexaploid in constitution it is possible that six genes represent the full range of hypersensitivity in the species. It is not known, however, how these genes compare with genes controlling hypersensitivity in other blight resistant species.

The above evidence illustrates the mutability of *Phytophthora infestans* and suggests that hypersensitivity as a form of protection is unreliable, particularly under epidemic conditions when mutant forms have opportunities for multiplication.

Distribution of races.—Although it appears that the potential number of different races is high—sixty-four on the basis of six R genes—less than half of that number have in fact been identified. The reason is that the identification of both genotypes and races are interdependent and must progress together.

Surveys in Britain have revealed the presence in the field of about a dozen races all identified by means of the original sixteen genotypes. Among them is one record of the appearance

of race (1, 2, 3, 4) in Northern Ireland (Proudfoot, 1959). It has not so far been found elsewhere in the British Isles.

Wide differences between the race populations of several countries have previously been observed (Black, 1957). During the last two years surveys of blight occurring in Brazil and Italy were made in co-operation with Professor A. Ciccarone and Dr B. Bastos Cruz. The isolates were tested at Pentlandfield.

The Brazilian isolates numbering sixty-five were collected in the state of São Paulo from both potato and tomato crops. The main features of the results were the prevalence of races (4) and (1,4) on potatoes and of races (0), (3) and (3,4) on tomatoes. The only other races in the collection were (1, 3, 4) and (2, 3, 4) and they were very limited in distribution. These results indicate that races (4) and (1,4) have a host preference for potatoes while races (0), (3) and (3, 4) are better adapted to parasitise tomatoes. It is probable that the blight population of Brazil has been affected to some extent by the importation of seed potatoes from Europe, particularly Holland and Germany, and the presence of race (2, 3, 4) could be explained in this manner since it is known to occur in these countries. None of the isolates proved to be pathogenic to the R_6 genotype.

The survey of blight in Italy was based on 183 isolates taken from both potatoes and tomatoes growing in widely scattered districts. The race population proved to be simpler than that in Brazil, potato crops being parasitised almost entirely by race (4) while tomato crops were predominantly affected by race (0). This evidence suggests that race (0) is primarily a tomato race while race (4) is specially adapted to potatoes. Races (3) and (3, 4) occurred on both potatoes and tomatoes, but only rarely, while races (2, 4) and (2, 3, 4) were each recorded once on potatoes.

By way of contrast, the blight population of Mexico which is the centre of origin of most of the blight resistant species of potato, is extremely complex. It is reported (Eide *et al.*, 1959) that the simpler races—*e.g.*, (0), (1), (2), (3), and (4)—predominate on *S. tuberosum* varieties whereas the more complex races occur mainly on wild species, and their derivatives, possessing combinations of R genes. Further, it has been found that no potato species remains entirely free from blight in that environment (Niederhauser, 1954). In other words, the race popula-

tion is so diverse that hypersensitivity as controlled by the R genes is of little or no avail.

These surveys illustrate the close correlation between the distribution of R genes and the distribution of the more complex races of the fungus. It is clear that the greatest variety of races is found in Mexico where the widest array of R genotypes is grown. Towards the other end of the scale is Italy where varieties with R genes are relatively uncommon and specialisation of the pathogen is correspondingly limited. In Britain and some West European countries, the race population appears to be intermediate in diversity and is probably correlated with the extent of the work and the progress made in breeding for blight resistance.

Origin of races.—It has been shown by Gallegly and Galindo (1957) that the blight population of Mexico comprises two inter-compatible groups of races and that intercrossing between the groups takes place freely under natural conditions. Thus the mechanism for genetic recombination, and hence for the production of new races, is at hand. The two compatibility groups were reported to be equally common in Mexico but no correlation between compatibility group and race appeared to exist. In these circumstances it is not surprising that physiological races are found in Mexico in great profusion.

The origin of new races in other parts of the world where only one of the compatibility groups has been found (Smoot, Gough and Gallegly (1957)) is difficult to explain. In the absence of sexual genetic recombination, the fungus still exhibits considerable plasticity. Many instances have been noted of changes from the simpler to the more complex races particularly those involving the gene R_4, for example :—

race	(0)	to race	(4)
,,	(1)	,,	(1, 2)
,,	(2)	,,	(2, 4)
,,	(3)	,,	(3, 4)
,,	(4)	,,	(1, 4)
,,	(1, 2)	,,	(1, 2, 4)

etc.

Mills and Peterson (1952) obtained five different races—viz., D, C, B, BD and BC—from race A by serial passage through

senescent leaves of the appropriate genotypes. According to the international nomenclature of races of *P. infestans* (Black *et al.*, 1954) this means that races (1), (2), (4), (1, 4) and (2, 4) respectively were obtained from race (0). It thus appears that with patience and suitable treatment, races pathogenic to all R genotypes could be produced.

Changes in pathogenicity, however, take place in the opposite direction with corresponding frequency. When the cultivation of R genotypes is discontinued in favour of *S. tuberosum* varieties the more highly specialised races tend to disappear and to be replaced by simpler forms. This has been observed, not only in the field but also in storage when tubers of commercial varieties lacking R genes were employed as media for maintaining cultures. In such circumstances a culture of :—

race	(1, 4)	reverted to race	(4)
,,	(1, 3, 4)	,, ,,	(3, 4)
,,	(1, 2, 3, 4)	,, ,,	(1, 3, 4)
,,	(1, 2, 4, 6)	,, ,,	(2, 4, 6.)

Such evidence serves to show that *P. infestans* is a remarkably adaptable organism. In the presence of R genotypes it can extend its host range but when R genotypes are replaced by ordinary varieties, simpler forms more in keeping with the host plants they encounter will reappear.

It would be interesting to know exactly how these changes take place. The fungus is probably heterocaryotic or it may have a parasexual cycle similar to that observed in *Fusarium*, *Aspergillus*, etc. It is also possible that genetic interchange between zoospores may take place. These are possibilities which merit further investigations.

Field resistance.—From the foregoing it is clear that R gene hypersensitivity cannot be relied upon as a permanent protection against the ravages of a parasitic fungus of such adaptability as *P. infestans*. When this seemed likely some years ago potato breeders began to concentrate more on field resistance which, although only a partial protection, appeared to be independent of racial specialisation in the pathogen. Field resistance may be defined as the degree of resistance exhibited by a plant towards all races of the parasite capable of causing more than a hypersensitive reaction on it. Field resistant plants have fewer lesions than susceptible plants. These lesions tend to be concentrated on the older leaves, to spread relatively slowly and to sporulate sparsely. A moderate degree

of field resistance has long been known in commercial varieties of *S. tuberosum* but it has been neglected for a time because of the initial promise of the R genes from *S. demissum*. However, the latter species is also an important source of field resistance factors and some of the seedlings bred from it have been found to possess both forms of resistance in good measure.

Much has still to be learned about the nature of field resistance and the various factors that contribute towards it. Since the range of variation from high field resistance to extreme susceptibility is more or less continuous the character is presumed to be controlled by a series of minor genes and to be inherited in polygenic fashion. The classification of field resistance is accordingly difficult to achieve and the lack of a convenient and accurate method of measurement is a handicap in its study and in the breeding of improved types.

Breeding specifically for field resistance has been in progress at Pentlandfield since 1954 when race (1, 2, 3, 4) became available for test purposes. This race gives a measure of field resistance in plants possessing any or all of the four R genes. The main source of field resistance has been *S. demissum* but other species such as *S. stoloniferum* and *S. simplicifolium* have also been involved.

In testing for field resistance the plants are sprayed with a spore suspension of the fungus and maintained in uniform conditions of high humidity for 24 hours to ensure that infection takes place. They are then exposed for a further 6 days to ordinary greenhouse conditions at 60°F, but are protected throughout the test from direct sunlight. At the end of the test the plants are classified as in Table 2, according to the type of the lesions and the amount of damage incurred.

TABLE 2

CLASSIFICATION OF FIELD RESISTANCE

Reaction group	Type of Lesion	Estimated area affected (%)	Description
1	Restricted	3	Highly resistant
2	Partly arrested	10	Fairly resistant
3	Retarded	30	Slightly resistant
4	Normally progressive	60	Normally susceptible
5	Rapidly progressive	100	Very susceptible

In this classification, field resistance is highest in Group 1 and lowest in Group 5. Most commercial varieties belong to Group 4 and consequently all members of Groups 1, 2 and 3 are superior to them in respect of field resistance. Despite the arbitrary nature of the scale, reasonable consistency may be attained with the aid of standard control varieties. This method of estimating field resistance is equally suitable for plants grown from tubers and for seedlings raised from the true seed.

The distribution of seedlings of six different progenies in the five categories of field resistance is shown in Table 3. These examples give some indication of the relationship between the field resistance of the parents and the distribution of such resistance in the offspring. In practice, the more resistant seedlings—*i.e.* groups 1 and 2 and some individuals in group 3—usually recover from the attack and produce a crop under greenhouse conditions. Thus the more promising resisters can be retained for further investigation and for breeding purposes.

TABLE 3

DISTRIBUTION OF FIELD RESISTANCE IN HYBRID PROGENIES

Reference number	Reaction group of parents	Distribution of seedlings in reaction groups					Total
		1	2	3	4	5	
3389	2 × 2	0	29	56	15	0	100
3319	2 × 3	0	8	70	19	3	100
2572	2 × 4	0	8	26	53	13	100
3308	3 × 3	0	8	18	70	4	100
2919	3 × 4	0	0	13	75	12	100
3331	4 × 4	0	0	2	79	19	100

Field resistant selections screened in this manner were submitted for test in Mexico through the generous co-operation of Dr J. S. Niederhauser of the Rockefeller Foundation. The results obtained showed that the degree of field resistance

exhibited in the laboratory tests was maintained under natural conditions in Mexico.

In a number of progenies, bred from *S. demissum* and other species, and tested with race (1, 2, 3, 4,) a proportion of plants reacted in hypersensitive fashion. When such plants were tested in Mexico they became blighted, some of them being described as very susceptible. Apparently they had been protected in the laboratory tests by a major gene—*e.g.* R_5 or R_6 conferring hypersensitivity to race (1, 2, 3, 4)—but had succumbed to other races prevalent in Mexico. The field resistance of some of the selections was, in fact, very low. These results emphasise the value of the Mexican tests, particularly as a guide in selecting true field resistant types for further breeding purposes.

In the list of seedlings submitted for test in Mexico, all five categories of field resistance were represented and some of the more resistant types yielded attractive crops. Accordingly the prospects of producing commercially acceptable varieties with a valuable degree of field resistance to blight appear to be good.

Summary

The identification of 16 different races of *Phytophthora infestans* is accomplished by means of a differential host series of 16 genotypes based on 4 R genes found in *Solanum demissum*. The R genes induce a hypersensitive response to infection with certain races of the fungus. Evidence is accumulating to show that *S. demissum* contains more than 4 such genes, probably 6, which in their different combinations would be capable of distinguishing 64 different races.

Blight surveys show that the number of races present in crops varies greatly from country to country. It tends to be low in *S. tuberosum* varieties and to increase as varieties possessing R genes are brought into cultivation. Thus the racial composition of blight populations fluctuates according to the R gene constitution of the potatoes grown.

A form of resistance known as "field resistance" gives a measure of protection against all known races of the fungus. It is present in *S. demissum* and other species and is inherited in polygenic fashion. By appropriate breeding methods it may be combined in high degree with essential commercial qualities.

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THE ROLE OF *SOLANUM VERNEI* BITT. ET WITTM. IN BREEDING FOR RESISTANCE TO POTATO ROOT EELWORM (*HETERODERA ROSTOCHIENSIS* WOLL.)

J. M. DUNNETT

Solanum ballsii was described from material collected by Hawkes and Balls in Northern Argentina in 1939 (Hawkes, 1944). Subsequently, Hawkes compared *S. ballsii* in the Commonwealth Potato Collection (C.P.C.) at Cambridge with *S. vernei* in the Erwin Baur Collection (E.B.S.) of the Max Planck Institute, Cologne, and referred (1956) to *S. vernei* subsp. *ballsii*, based on *S. ballsii*. The E.B.S. stock of *S. vernei* was acquired from Brücher, who rediscovered the species in Northern Argentina in 1950, thirty-six years after the original collection by Wittmack (Goffart and Ross, 1954). Brücher (1957) found no evidence to support the retention of *S. ballsii* as a sub-specific name.

S. vernei is not a cultivated species but is a probable ancestor of cultivated potatoes of the " *S. andigenum* group " (*S. tuberosum* L.) according to Brücher (1954).

Ross and Baerecke (1951) detected " incubation resistance " to *Phytophthora infestans*, the late blight fungus, resistance to potato virus Y and resistance to frost in *S. vernei*.

The resistance of S. vernei to potato root eelworm.—Ellenby reported (1948) that the C.P.C. stock of *S. vernei* was resistant to potato root eelworm. Larvae were present in the roots of the plants exposed to infection, but even immature cysts were very rare. Ellenby's findings were confirmed in the U.S.A. by Mai and Peterson (1952) who worked with material obtained from Cambridge. Macdonald (1956), also working in the U.S.A., recovered brown cysts from *S. vernei* of unspecified origin. Goffart and Ross (1954) and Rothacker (1957) found the E.B.S. stock of *S. vernei* to be outstandingly resistant, although the plants were not always free from cysts. Rothacker (1957) listed a " susceptible " clone of *S. vernei*. Williams (1958) compared *S. vernei* with resistant clones bred from *S. tuberosum* subsp. *andigena* and found that the plants of *S. vernei* produced a much less active root diffusate, and were invaded by fewer larvae, of which very few developed past the second or invasive

stage, and adults of both sexes were absent. Dunnett (1957, 1958) found that C.P.C. and E.B.S. stocks of *S. vernei* remained virtually free from cysts and certainly free from cysts past the white stage of development, when infected by either the Boghall or Duddingston populations of the eelworm maintained at the Scottish Plant Breeding Station. These populations differed in pathogenicity towards plants possessing the resistance of subsp. *andigena* C.P.C. 1673, the Duddingston population being the aggressive population. Jones (1957) reported *S. vernei* resistant to thirty-seven populations, against most of which the subsp. *andigena* type of resistance was known to be more or less ineffective. Dunnett (1958) tested P and H 351, a new collection of *S. vernei* made by Peterson and Hjerting and observed no cyst formation on six clones infected by the Duddingston population.

Therefore, the resistance of *S. vernei* is equally effective against the two types of pathogenicity detected to date in naturally occurring populations of *H. rostochiensis*. Susceptible clones occur exceptionally, although none have been found in the C.P.C. stock of the species. The cysts which arise occasionally on the resistant clones are usually small and white: conversely the " occasional " cysts found on resistant clones bred from subsp. *andigena* are usually fully grown and contain eggs. Recent work (Ellenby, 1957; Fassuliotis, 1957; Williams, 1957) has shown that fertilisation is essential for egg production. It is significant that the development of both sexes is equally inhibited in *S. vernei* whereas an excess of males over females matures in resistant plants bred from subsp. *andigena* (Jones, 1954; Williams, 1958).

Inheritance of resistance.—Goffart and Ross (1954) postulated polygenic inheritance of resistance in crosses between colchicine-induced tetraploid clones of *S. vernei* and *S. tuberosum*, with a tendency for resistance to be dominant. Rothacker (1959) took the same view and suggested that resistance might be due to one or two genes with modifiers. Goffart (1957) mentioned a breeding line stemming from *S. vernei* which was discontinued because resistance decreased as breeding progressed. At Pentlandfield, the hybrids between *S. vernei* (4n) and *S. tuberosum* retained a high level of resistance to the Duddingston population but none remained free from cysts. The infectivity of the Duddingston population declined to four-fifths of the initial level of infectivity in the culture

medium in which the most resistant hybrid progeny had grown.

A comparison of breeding lines.—Resistance to potato root eelworm which may be useful for breeding purposes occurs in the following species of potato indigenous to South America : *S. tuberosum* subsp. *andigena*, *S. vernei*, *S. famatinae*, *S. multidissectum*, *S. neohawkesii*, *S. sanctae-rosae*, *S. megistacrolobum*, *S. raphanifolium*, *S. capsicibaccatum*, *S. microdontum* and *S. polyadenium* (Anon., 1959; Ross, 1958). All are diploid species excepting the tetraploid subsp. *andigena*. The evaluation of the resistance in these species is still incomplete because only a few breeding lines can be pursued simultaneously. Currently, subsp. *andigena* C.P.C. 1673 and *S. multidissectum* are the principal sources of resistance in breeding at Pentlandfield.

The level of resistance of *S. tuberosum* subsp. *andigena* C.P.C. 1673 to the Boghall population was specified (Anon., 1959) as that which resulted in 90 per cent of the plants remaining cyst-free on the basis of soil-ball inspection after exposure to infection in 4-inch pots containing a cyst-infested culture medium of 40 eggs/gm. infectivity. This level of resistance was regarded at Pentlandfield as a "standard" level of resistance to aim at in new varieties of potato. *S. multidissectum* possessed standard or above standard resistance to the Duddingston population and a dominant gene appeared to be mainly responsible for the resistance (Dunnett, *in lit.*). The resistance of subsp. *andigena* C.P.C. 1673 was controlled by a dominant gene designated H (Toxopeus and Huijsman, 1953). The Boghall and Duddingston populations were aggressive towards plants possessing the resistance of *S. multidissectum* and subsp. *andigena* C.P.C. 1673, respectively.

The polygenic resistance of *S. vernei* is a disadvantage for breeding purposes because hybridisation with commercial varieties of *S. tuberosum*, the first step in breeding, results in some loss of resistance throughout the F_1. It is necessary to select and intercross the more resistant seedlings in the more resistant progenies after crossing with *S. tuberosum* if a variety of potato possessing a standard level of resistance derived from *S. vernei* is the objective. There is a better prospect of incorporating the resistance of subsp. *andigena* C.P.C. 1673 and *S. multidissectum* in commercial varieties of potato because segregates with a standard level of resistance occur in the backcrosses to *S. tuberosum*.

A second disadvantage of *S. vernei* is the fact that the resistant plants do not stimulate a high rate of larval emergence from cysts in the soil and therefore lack an essential feature of a successful " trap crop " for potato root eelworm. Plants having the resistance of subsp. *andigena* C.P.C. 1673 possess this " trap cropping " property to a much higher degree.

Despite these disadvantages, *S. vernei* may still play an important role in breeding for resistance to potato root eelworm. The scope of the resistance in relation to variation in the pathogenicity of *H. rostochiensis* may be broader than that of subsp. *andigena* C.P.C. 1673 and *S. multidissectum*, since no populations aggressive towards *S. vernei* are known at present. Until aggressiveness towards *S. vernei* can be demonstrated, the species must be regarded as almost outside the host range of *H. rostochiensis* and the resistance may be proof against the whole range of possible variation in the pathogenicity of the pest. This kind of resistance would be very valuable and comparable in significance to the polygenic " field resistance " of *Solanum demissum* to *Phytophthora infestans*, the fungus causing late blight of potatoes. Even part of the resistance of *S. vernei* combined with resistance to particular strains conferred by dominant genes derived from subsp. *andigena* C.P.C. 1673 and *S. multidissectum* might delay the evolution of new aggressive strains of the eelworm.

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NOTES ON THE BOTANICAL CHARACTERS OF SOME RAPE TYPES

V. McM. Davey

The West of Scotland Agricultural College had undertaken an investigation into the uses and productivity of certain types of rape and kale, and the Scottish Plant Breeding Station was invited to contribute some information about the botanical characters. A trial, and observation plots were therefore laid out at Pentlandfield in 1959. The seed supplied by the College consisted of three varieties of rape (*Brassica napus* L.), viz. giant rape, dwarf rape and " Margo " rape, obtained from local seedsmen ; the first two were British varieties, while Margo was of Dutch origin. The Station obtained for comparison three certified varieties of rape seed from New Zealand, and since it may help to clarify the situation, a short account of how these varieties came into being will first be given.

New Zealand certified rape varieties

During and after the first World War, New Zealand was dependent upon imported rape seed. As much as 3,420 cwt. of seed was bought from Britain in 1931, but during the war supplies had been very severely restricted. Moreover, certain diseases were now found to be seed-borne, and it was thought that dry rot of swedes was being spread through New Zealand in this way. The home production of rape seed was therefore advocated by Hadfield (1933a, part I), who outlined a production technique and suggested a certification scheme (1933a, part II). Confusion of nomenclature in New Zealand had earlier led Hadfield (1931) to survey the characters of a number of commercial varieties, and he defined the following three types :—

" Type 1 is a large-leaved Giant rape giving a greater initial bulk of feed than the other types, but not the recovery of Type 2.

" Type 2 is of a dwarf habit, producing a dense crown of many leaves. Its initial yield is not as great as that of Type 1, but recovery is better.

> " Type 3 is open in the crown and flat, the leaves bluish-green and much dissected. The initial yield and recovery are very much less than in Types 1 and 2. It runs to seed very early, and present indications are that it is a very undesirable type."

Hadfield concluded that the trade names under which rape was then sold were no indication of its type, and his table showed that out of twenty-three varieties, sixteen were called Broad-leaved Essex, and though most of these were of type 2, several were classed as types 1 and 3. The confusion may not be so great in Britain at the present day, but it is not always clear to which type the terms " broad-leaved " and " Essex " are being applied.

The outcome of Hadfield's survey was that breeding was commenced under the auspices of the New Zealand Department of Industrial Research, and very soon strains of types 1 and 2 were ready to be multiplied under a certification scheme (Hadfield, 1933b). The Giant Rape was true type 1, while the " Broad-leaved Essex " was type 2 with a dash of giant " blood," to improve the initial yield. Selection for resistance to clubroot was started by the plant pathologist, Dr J. G. Gibb, and continued by R. A. Calder (Lobb, 1951), but the first introduction of a resistant strain to the public seems to have been in 1949 when Claridge defines the three strains, as they were then termed, as follows :—

1. *Certified Giant.*—A high-producing type showing very little recovery, which has been developed by plant selection.
2. *Certified Broad Leaf Essex.*—A type developed by hybridisation which gives a good initial yield and has the ability to recover after grazing.
3. *Certified Clubroot Resistant.*—A strain of rape of the Giant type showing pronounced resistance to attack by clubroot disease.

Seed of the Clubroot Resistant was still in short supply in 1951 when Lobb recommended its use for land where rape growing had been abandoned because of clubroot. He stresses that the strain is highly resistant, but that no immunity is claimed.

Field trials

Since the agronomic aspects were being studied by the West of Scotland Agricultural College, it was unnecessary to conform with agricultural practice when laying out trials and observation plots. During the last few years, varieties of rape have been sown at Pentlandfield on various dates in late spring and summer, and it has been found that the April and May sowings generally give satisfactory plants stands and vigorous growth, whereas June and July sowings have usually failed. The main sowings in 1959 were therefore carried out on the 5th and 6th of May, and only one " catchcrop " sowing was made in July. In agricultural practice, rape plants are grown crowded together and get insufficient space for full vegetative development. Spacing of plants was indicated for an examination of their potential growth, and in the main trial for spaced plants, the seed was sown on 5th May in dibble holes 10 in. apart along 26 in. ridges ; thereafter the trial was cultivated like a swede crop. Each variety had five plots in randomised blocks, and each plot had a potential stand of 86 plants. A group of 10 plants was taken from inside each plot for weighing and measurement, leaving about 350 per variety upon which to observe bolting.

A seed-bed was also sown on May 5th-6th, at the rate of 100 seeds per 8-ft. strip. It was not practicable to lay out a replicated trial and the four strips of each variety were intended to provide seedlings for transplanting in June. Two rows were indeed used for this purpose, but two were left growing and one of these was cut for measurement, the other observed for flowering. Two blocks of land were used for transplants spaced at 24 in. × 24 in., and forty or fifty seedlings of a variety were planted in each block. A row of 10 plants was cut from each plot in August for weighing and measurement, the rest being observed for bolting. Broadcast sowings of the three New Zealand rapes were made on 14th July, and sowings were also made in boxes of soil for the testing of resistance to clubroot.

The six varieties of rape are each denoted by the same letter throughout the trials, viz. :—

Seed obtained locally	*Seed from New Zealand*
A. Giant (British)	B. Certified Giant
C. Dwarf (,,)	D. ,, Broad-leaved Essex II
E. Margo (Dutch)	F. ,, Clubroot resistant

Bolting behaviour.—Leafy rapes, as distinct from oil-seed forms, are bred to make rapid vegetative growth and not to bolt during the autumn after they have been sown in summer. When sown earlier, however, bolting tendencies may be revealed, and these differ with the variety as may be seen from Table 1.

TABLE 1

BOLTING TENDENCIES IN RAPE SOWN ON 5-6 MAY

Type	1 Giant rape		2 Dwarf or B.L. Essex			Clubroot resistant
Country of origin Variety	U.K. A	N.Z. B	U.K. C	N.Z. D	Holland E	N.Z. F
Spaced plants, 10″ × 26″						
Number of plants	371	368	352	377	338	376
8 August) % bolted	3.8	41.5	2.8	0.3	3.3	96.8%
95 days) % in flower	0.3	1.4	1.7	0.3	1.8	9.3%
28-30 August) % bolted	8.1	99.5	18.5	4.8	21.9	100.0%
115-7 days) % in flower	4.3	34.9	12.5	2.9	16.6	89.4%
Transplants, 24″ × 24″						
Number of plants	63	52	63	73	73	74
19 September) stem length	10″-18″	24″	12″	6″	short	long
136 days) % in flower	5	29	17	0	21	69%
40-50 *plants in 8′ drill*						
19 September) stems	long	bolted	12″	6″	short	in pod
136 days) % in flower	0	most	½	0	1	100%

The New Zealand giant, variety B, bolted fairly completely in the spaced-plants trial and in the crowded seed-bed strip, though only 29 per cent of the transplants flowered. On the other hand, the New Zealand type II, variety D, showed the least bolting of all. Variety A, the British sample of giant type, was much less affected by bolting than its New Zealand counterpart, and less also than the British and Dutch dwarf types C and E. These dwarfs were relatively slow to start but eventually about 20 per cent bolted in the spaced trial and the transplants; very few dwarfs bolted in the seed-bed,

TABLE II

YIELD OF LEAF AND STEM IN PLANTS OF RAPE SOWN 5-6 MAY

Type of rape	1 Giant rape		2 Dwarf or B.L. Essex			Clubroot resistant
Country of origin / Variety	U.K. A	N.Z. B	U.K. C	N.Z. D	Holland E	N.Z. F
Spaced plants, 10″ × 26″						
4-10 August) leaf weight	621	719	635	684	623	594 g.
91-97 days) stem weight	103	262	91	84	82	189 g.
ratio leaf : stem	6.1 : 1	2.8 : 1	7.0 : 1	8.3 : 1	7.5 : 1	3.1 : 1
Transplants, 24″ × 24″						
12 August) leaf weight	1370	1610	1355	1345	1360	1430 g.
98 days) stem weight	315	530	245	250	265	360 g.
ratio leaf : stem	4.3 : 1	3.0 : 1	5.5 : 1	5.4 : 1	5.1 : 1	4.0 : 1
25 August) leaf weight	1700	1675	1870	1510	1575	1380 g.
111 days) stem weight	475	860	500	490	430	575 g.
ratio leaf : stem	3.6 : 1	1.9 : 1	3.7 : 1	3.1 : 1	3.7 : 1	2.4 : 1
40-50 *plants in 8′ drill*						
16 August) leaf weight	174	233	240	162	248	188 g.
102 days) stem weight	36	135	39	30	33	100 g.
ratio leaf : stem	4.8 : 1	1.7 : 1	6.2 : 1	5.4 : 1	7.5 : 1	1.9 : 1

however. The most active bolter was the clubroot resistant F. As early as 78 days after sowing, on 22nd July, plots of F were recognisable because the foliage was carried higher than that of the surrounding crops. By 95 days nearly every plant of F in the spaced trial was so shot that the terminal flower buds were plainly visible, and 9 per cent had flowered. The crowding of plants in the seed-bed strips seems to give more contrasting results than are found in the other spacings ; while the dwarfs were hardly affected, the plants of variety F had all run to pod. It should be stressed, however, that all these results followed sowing in early May. In the broadcast plots where the three New Zealand varieties were sown on 14th July, no bolting was observed even in January. All the varieties had the same flower colour, bright lemon yellow.

Several tones of yellow occur in rape (Sylven, 1927), and a pale-yellow flower sometimes characterises a variety.

Production of leaf and stem.—The results of weighings are given in Table II. The trial of spaced plants at the top of the Table can be treated statistically, the transplants and seed-bed records are put in for comparison. Ten plants were taken from each plot of the spaced plant trial; two replications being sampled on 4th August, two on 7th and one on the 10th of that month (91-97 days' growth). Outside plants were rejected.

Each plant was weighed separately after the spindle had been removed; then the leaves were stripped off and the stems weighed and measured. The mean leaf weight varied from 719 g. in the New Zealand giant, B, to 594 g. in clubroot resistant F, but so large were the errors that this extreme difference was not significant. On the other hand, the differences in stem weight were more accurately determined, and those exceeding 36 g. may be regarded as significant (P = 1 per cent). Thus the stems of New Zealand giant, B, weigh more than those of any other variety. The clubroot resistant, F, comes second and is significantly higher than all except B. The British giant, A, with a stem weight of 103 g., is not significantly greater than the dwarf types. Mainly on account of stem, the gross plant weights (*i.e.* stem + leaf) also show some significance; a difference of 214 g. having a probability of 1 per cent. Variety B with 981 g. again heads the list, being significantly greater than A, C and E, but not D or F. There are no other significant differences. Differences in yield were in fact due to bolting in varieties B and F. When these weighings were made the stems were quite soft, and there was no reason to suppose that they were less edible than the leaves. Only the spindle is woody, and this hardly projects above the surface of the soil. From an agronomic viewpoint, however, the rape in this trial was past its prime, the leaves were turning dark green and new ones were not growing fast to replace them. Shortly afterwards an unusually early and severe attack of mildew was to cover the foliage. All the rape suffered, possibly the dwarfs showed more mildew or rather the upper leaves of the giant and clubroot resistant varieties remained green longer, but no real differences could be distinguished. After the drought ended there was a period when the leafage looked fresh again before entering the winter dormancy. It

may be mentioned in passing that while rape and swedes suffered badly from mildew, Canson and other kales were quite unaffected.

The other weights shown in Table II illustrate extremes due to conditions of growth. The figures for transplants are the mean weights of ten plants cut and weighed individually as in the spaced trial. Despite the check due to transplanting, these had been able to grow luxuriantly. The cutting made on 12th August shows leaf weights twice as great as those of the plants spaced at 10 in. The giant and clubroot varieties have about twice as much stem, but the three dwarfs are quite two and a half times as heavy as in the spaced trial. The other transplants which were cut thirteen days later showed heavier weights still, but judging by the ratios the stems were increasing faster than the leafage.

Finally, there are the seed-bed strips of crowded plants whose stems weighed merely an ounce or two. The bolted New Zealand giant, B, and clubroot resistant F, had heavier stems than the others as might be expected, but the numbers of plants per row, which ranged from 57 to 37, invalidate the comparisons of varietal differences. All values are somewhat enhanced by the inclusion of end plants adjacent to paths.

Stem development.—The bolting observations and stem weights have already built up some picture of the different responses of the varieties in the three environments. Measurements of stem length show another aspect, and may also add some information about variability. In Table III data obtained from the measurement of stems of the spaced plants are compared with those of plants growing in the crowded seed-bed strips. It may be admitted that the first data refer to groups of plants scattered over a sixth of an acre, and not sampled on the same day, but though one block of the spaced trial lacked vigour, there was no significant difference between replications. Comparing mean lengths, the two groups appear to have grown to about the same heights. New Zealand giant, B, is tallest, closely followed by the clubroot resistant, F. The mean length of British giant, A, is not much more than those of the dwarfs of which the Dutch Margo appears to be shortest.

Observation suggests that the dwarf-type plant remains relatively short-stemmed unless and until it is activated into bolting, whereas the stem of the giant lengthens to some

TABLE III

Length of stem in spaced and close-growing populations

Type of rape	1 Giant rape		2 Dwarf or B.L. Essex			Clubroot resistant
Country of origin Variety	U.K. A	N.Z. B	U.K. C	N.Z. D	Holland E	N.Z. F
50 spaced plants, 10″ × 26″ (4-10 Aug. 91-97 days)						
Mean stem length in cm.	22.6 ± 1.53	57.2 ± 3.35	18.1 ± 3.26	14.7 ± 0.63	11.1 ± 0.82	54.7 cm. ± 1.63
Standard deviation in cm.	11	24	23	4	6	11 cm.
Standard deviation % of mean	48	41	128	30	52	21%
Number in flower	0	2	3	0	0	*
Range in cm. maximum in flower	..	150	150	..	..	* cm.
Range in cm. maximum, no flowers	58	95	30	34	28	87 cm.
Range in cm. minimum	8	25	0	8	1	30 cm.
Plants crowded in 8′ drill (16 Aug. 102 days)						
Mean stem length in cm.	20.9 ± 1.56	65.4 ± 5.06	15.8 ± 2.77	15.9 ± 1.20	9.2 ± 1.04	61.4 cm. ± 3.65
Standard deviation in cm.	11	32	18	9	6	24 cm.
Standard deviation % of mean	53	49	114	57	69	39%
Number of plants	51	40	42	57	37	42
Number in flower	0	3	1	0	0	4
Range in cm. maximum in flower	..	165	120	..	..	150 cm.
Range in cm. maximum, no flowers	55	95	30	51	34	95 cm.
Range in cm. minimum	5	20	4	5	2	30 cm.

* Most in flower bud stage

extent even when the foliage remains typically vegetative. To see whether this impression can be confirmed, some data on variation and flowering have been included in Table III. Comparing the giants and dwarfs, varieties B and C, in which some flowering occurred, had larger standard deviations than

A, D and E. In the dwarf C, the standard deviations exceeded the mean values. The non-flowering plants of the three dwarf varieties range from sessile to a top level of about 35 cm. One stem in the crowded population of variety D measured 51 cm., but the next longest were three at 35 cm., and as variety D showed a few plants in flower elsewhere (cf. Table I) it is not improbable that the 51 cm. plant was caught in the act of bolting. Compared with this low-growing habit of the dwarfs, the New Zealand giant, B, is obviously long-stemmed, but the British giant, A, is not quite so distinct, stems range up to about 55 cm., but there are short ones in both populations.

The clubroot resistant variety, F, is said to be of giant origin, and it is certainly more ready to lengthen stem than either giant. In the crowded populations, F and B show similarity in mean and range, but there is a peculiar situation in the spaced population of variety F. A stage of bolting has been reached at which all the plants are long-stemmed, between 30 and 87 cm., with flower-buds showing, but terminal racemes as yet undeveloped. In consequence the standard deviation is small, both in actual value and in proportion to the mean, as may be seen by comparing the values with those of the spaced population of the giant B.

Development in broadcast plots.—Seed of the three New Zealand varieties was sown broadcast in large adjoining plots on 16th July. Since these varieties had shown distinct differences in the earlier sowings, it was of interest to see how the stems developed under a more usual set of conditions. Two samples were taken from each plot on 30th September and 15th October respectively, and the data are summarised in Table IV. It should be emphasised that no flowering or bolting was found then, nor, indeed, did any appear before January. The samples comprised all plants within a square grid, $4' \times 4'$ in area, and the plant stands differed to an extent which certainly affected some of the gross plant weights, though perhaps the leafage more than the stems.

The first sample of variety D had a sparse stand and the second sample of F was relatively dense; the leaf and gross weights of these two are high and low respectively. The stems of D, the New Zealand B.L.E. II, weigh less than those of either giant, B, or clubroot resistant, F, and they are barely half the length. This is seen from the mean values and also from the ranges. There are a few undersized plants in every

TABLE IV

SAMPLES CUT FROM PLOTS SOWN BROADCAST ON JULY 14TH

Type of rape	N.Z. giant		N.Z. B.L.E. II		N.Z. clubroot resistant	
Variety	B		D		F	
Date of cut	30 Sept.	15 Oct.	30 Sept.	15 Oct.	30 Sept.	15 Oct.
Days of growth	78	92	78	92	78	92
Plant weight						
gross	109	126	118	96	111	92 g.
leaf	93	103	108	84	95	75 g.
stem	16	24	10	12	16	17 g.
leaf : stem	5.8 : 1	4.4 : 1	10.3 : 1	7.2 : 1	6.1 : 1	4.4 : 1
Plant stand						
per sq. yard	29	26	21	26	25	34
Stem length						
mean	15.4	20.3	8.1	9.7	16.0	19.4 cm.
	± 0.72	± 0.99	± 0.39	± 0.37	± 0.81	± 0.62
S.D. in cm.	5.2	6.8	2.4	2.6	5.4	4.8 cm.
S.D. % mean	33	84	30	27	34	25%
range maximum	28	38	15	16	27	30 cm.
range minimum	5	7	5	5	5	6 cm.

cut, giving a minimum stem length of about 5 cm., but whereas the longest of variety D was only 16 cm., stems reached 38 cm. in the later cut of the giant. It should be mentioned that these plants were cut with a pruning knife about 2 cm. above the spindle.

Resistance to Clubroot.—No clubroot was found in the field where the rape was grown, and no infected plots are available at Pentlandfield, so a trial was carried out with seedlings growing in boxes to see whether the New Zealand clubroot resistant variety, F, would exhibit resistance superior to the other rapes. Seed of the six rape varieties, two samples of thousand-headed kale and the disease-resistant turnip, "The

Bruce," was sown on 19th May in twelve boxes containing sterilised, limeless, potting compost. The seed was sown in dibble holes and thinned so that seven plants of each variety occupied one of nine randomised cross rows in each box. On 19th June an aqueous extract from highly infected soil was poured evenly over the contents of eleven boxes. The twelfth box was left uninfected, and none of its 63 seedlings showed any clubroot, so that there was no infection in the original compost. Counts of the plants in the eleven boxes were made on 21st July, and are summarised in Table V.

TABLE V

RESISTANCE OF SEEDLINGS TO CLUBROOT DISEASE

Description		Total	Free	Showing disease	
				slight	serious
Clubroot resistant	F	77	77	0	0
N.Z. B.L.E. II	D	77	60	17	0
Dutch Margo	E	75	54	19	2
British dwarf	C	77	52	23	2
British giant	A	77	50	24	3
N.Z. giant	B	76	47	21	8
Bruce turnip	J	77	77	0	0
1000-headed kale	H	77	69	8	0
Canson kale	G	77	69	6	2

Two boxes contained no diseased plants so that the *Plasmodiophora* may have met with adverse soil conditions. However, even including these negative results, the difference between clubroot resistant rape, F, with no disease showing, and the other rape varieties is obvious. The latter showed susceptibility, from 22 per cent in variety D to 38 per cent in B, but distinctions should not be drawn between them because, given susceptibility, the wet end of a sloping box might increase the number of casualties in one or two varieties.

The Bruce turnip control showed complete resistance, and the thousand-headed kales had about 10 per cent slightly damaged. This kale is usually regarded as relatively resistant.

Discussion.—The New Zealand investigations, of which a short account has been given above, suggest that the varieties of rape grown in this country can be classified either as type 1 or 2, and that the two certified varieties are characteristic representatives of these types. This is certainly the case, the giants are similar in appearance, while the British dwarf and Margo closely resemble the New Zealand Broad-leaved Essex. The chief distinction lies in the degrees of bolting that can be induced in varieties by early sowing. Before the war, samples of New Zealand types 1 and 2 were sown in the swede break at Corstorphine for comparison with "rogue" plants found in swede crops and sent in for identification. It was then noted that the giant always had long stems and some plants in flower in autumn, while the type 2 samples were very resistant to bolting. It is interesting to find the same distinction now, but it is not clear whether the characters have any practical value, or have become accidentally bound up in the inheritance of these varieties. The New Zealand clubroot resistant variety, which is said to have been selected from a French variety of giant type, is distinctly susceptible to bolting when sown early, but this is unlikely to affect summer sowings, except that the stems may be somewhat lengthened. The British giant is less susceptible to bolting than its New Zealand counterpart, while the British dwarf and Margo bolt more than the New Zealand B.L.E. There were not enough samples of the numerous varieties grown in Britain to know whether these tendencies are general or merely characteristic of these particular varieties.

So far as is known no British variety of rape can claim the degree of resistance to clubroot exhibited by the special New Zealand variety. When tested in the strongly infected field plot at Corstorphine in 1950, all the rape plants showed nodules, but clubroot resistant rape carried fresh green leaves when the dwarf and giant controls were practically dead. The box test described above was not very severe, because none of the resistant turnips showed disease, but it was sufficient to affect a considerable number of kale plants.

Under broadcast conditions and in crowded rows the axillary buds of rape plants have little or no chance to develop until

the plant has been cut down or grazed. The widely spaced transplant, however, is able to develop side shoots after a short period and irrespective of whether it is inclined to bolt or remain vegetative. The closer spaced plants of the trial held an intermediate position, and showed diversity in development when cut and examined. It was not possible, however, to distinguish varietal differences because development was complicated. There was firstly a general development affecting the lowest buds most and the upper ones to a less degree, but superimposed on this there might be the favoured growth of one or a few branches. The typical dwarf with its short stem has many buds near ground level, and this, no doubt, helps to give it better recovery than the giant after grazing.

A second question set by the West of Scotland Agricultural College was the comparison of Canson Kale with rape. Canson is a form of thousand-headed kale (*B. oleracea acephala* L.) characterised by a leafy and relatively uniform habit of growth. In recent years it has been used on a considerable scale in parts of Wales and England, being sown broadcast for grazing in autumn and winter. As such it comes into competition with rape, but the growth rates of the two are so different that sowings made on the same date cannot be compared at the same time. The data from treatments of Canson and ordinary thousand-headed kale have, therefore, been omitted from the Tables, except in the clubroot test. The kales carried relatively few leaves when weighed, but later when the rape had become dormant they were yielding heavily. Kale is resistant to clubroot and mildew; it has no tendency to bolt before December but it possesses very little power of recovery after grazing compared with the rapes.

Summary

Characteristics of giant and dwarf rape were examined in several conditions of spacing, using certified varieties from New Zealand as controls. Bolting tendencies differ within types; when sown in early May, New Zealand giant rape, type 1, bolted considerably, but a British variety of giant was not much affected. Representing type 2 or dwarf rape, the New Zealand Broad-leaved Essex was highly resistant to bolting, while a British dwarf variety and the Dutch Margo,

each had about 20 per cent of bolted plants in 117 days. Differences in leaf weight were insignificant, but the New Zealand giant had heavier stem weights than the others. Plants of dwarf varieties remained short-stemmed unless they bolted, but giant plants showed some lengthening of stem while their foliage was still vegetative.

A New Zealand clubroot resistant variety showed more rapid and complete bolting than any of the above, and its long stems were heavier than those of dwarf rape. None of its plants had nodules when grown in boxes of clubroot infected soil; whereas 22 to 38 per cent of plants in the other rape varieties showed disease.

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LEAF LENGTH VARIATION IN *BETULA PUBESCENS* EHRH.

F. J. W. England

Introduction.—In studies of variation in wild populations, it is often a fairly simple matter to demonstrate significant differences between populations but a much more difficult matter to link these differences to variation in the natural environment. In the investigation reported here an attempt has been made to demonstrate a connection between variations in leaf length in *Betula pubescens* Ehrh. and certain measures of temperature variation in the environment. In addition some observations were made to determine to what extent phenotypic variation in leaf length in *B. pubescens* is genetically determined.

Material and methods.—The material on which the greater part of the investigation is based consists of two collections of leaves of *Betula pubescens* Ehrh., one made in 1950 and the other in 1953. In both years the collections were made during July. The leaves themselves were not preserved but carbon paper rubbings were made of them and all measurements were made on these rubbings.

Details of the populations are given in Table 1. The 1950 collection includes nineteen populations and was confined to the central Highlands of Scotland. It was made along a transect running from the mouth of the River Spey westward to Newtonmore and from there westward again to Arisaig. An attempt was made to take samples at five-mile intervals; this was not always possible since although birch is abundant in the eastern part of the transect much of it is *B. pendula* (Roth.). In this year only one leaf from each tree was taken and for each population approximately fifty trees were sampled.

The 1953 collection consists of eighteen populations from a rather wider area and includes four populations from England. Only one population, that at Newtonmore, was included in both years and no attempt was made to sample the same trees on both occasions. In 1953 two leaves were taken per tree and in general fifty trees were sampled from each population.

In both years the leaves sampled were those from sterile short shoots and the longest leaf on the shoot was taken for

measuring; the two leaves from each tree in 1953 were taken from different short shoots.

Various measurements were made on the leaf rubbings but in this paper we are only concerned with leaf length. It may, however, be mentioned that most other simple measurements are so highly correlated with length as to yield little extra information. This is true, not surprisingly, of leaf breadth; but also of basal angle and various estimates of number of teeth between successive veins. It should be emphasised that this comment applies to population means and not necessarily to the within population variation.

The temperature measurements were derived from the Monthly Weather Report of the Meteorological Office. In estimating any temperature measurement for the site of a birch population the records for one or several nearby meteorological stations were used as a basis. These records were weighted to allow for altitude, it being assumed that a 1°F fall in temperature occurs for every 300 feet increase in altitude.

From the mean monthly temperatures derived as above an estimate of the length of growing season was made. The definition of the growing season adopted by Anderson and Fairbairn (1955) was accepted for this purpose. Following Hagen, these authors define the growing season as being ". . . that period of time—in number of days—which lies between the first day of spring, whose mean temperature rises to 45°F, and the first day in autumn, whose mean temperature fails to reach 45°F."

For the sake of convenience, the various temperature and growing season measurements referred to later are listed below:—

A_1 length of average growing season in days.
A_2 average annual mean temperature (average mean of daily mean temperature).
A_3 average date of start of growing season.
C_3 "current" date of start of growing season (*i.e.* date of start of growing season in the year in which the leaves were collected, either 1950 or 1953).
A_4 average May temperature (average mean of daily means).
C_4 current May temperature.
A_5 average accumulated growing season temperature.
C_5 current accumulated growing season temperature.

TABLE 1

LOCATIONS AND ALTITUDES OF *B. PUBESCENS* POPULATIONS

Pop. No.	Name	OS	REF.	Mean Alt. Feet	Leaf Length mm.	Year
1	" Speymouth "			250	33.0	
2	Grantown-on-Spey	NJ	0328	870	28.3	
3	Boat of Garten	NH	9520	800	33.6	
4	Aviemore	NH	9015	880	27.6	
5	Kincraig	NH	8606	900	26.4	
6	Kingussie	NN	7799	900	28.0	
7	Newtonmore	NN	6896	1,300	29.9	
8	Loch Laggan	NN	5791	1,300	30.3	
9	" Lagganside "	NN	4789	1,400	36.0	1950
10	Roybridge	NN	3281	640	33.1	
11	Spean Bridge	NN	2381	410	36.0	
12	Achnasaul	NN	1787	410	35.1	
13	Corpach	NN	1280	250	34.3	
14	Fassfern	NN	0279	700	32.3	
15	Kinlocheil	NM	9480	100	37.8	
16	Ranochan	NM	8482	300	34.9	
17		NM	7483	250	35.1	
18	Coille Ropach	NM	7185	200	36.2	
19	Rhill	NM	6685	100	34.6	
21	Okehampton	SX	5797	500	42.2	
22	Hackness	SE	9591	700	42.2	
23	Girvan	NX	2298	200	39.1	
24	Borden	SU	8036	150	42.1	
25	Arisaig	NM	6687	100	40.3	
26	Tomich	NH	3228	700	36.1	
27	Newtonmore	NN	6896	1,300	33.3	
28	Penicuik	NT	2358	700	37.3	
29	Bridge of Brown	NJ	1519	1,100	36.9	1953
30	Glen Shiel	NG	9417	400	37.8	
31	Loch Carron	NG	9141	200	38.3	
32	Cluanie	NH	0712	1,000	32.4	
33	Tomatin	NH	8129	1,000	31.3	
34	Achnasheen	NH	2060	800	35.2	
35	Kinlochleven	NN	1763	100	39.1	
36	Lin Gill	SD	8078	1,200	41.8	
37	Achanalt	NH	2762	600	35.2	
38	Invergarry	NH	2902	400	36.9	

In addition to the two leaf collections made in the wild, measurements of leaf length were also made on six-year-old trees grown in the experimental garden at Pentlandfield from seed collected from seven populations in the wild in 1951. In the garden each population was represented by between fifty and seventy-five trees grown in a random block of four replications (originally each population contained one hundred trees but there has been considerable mortality since planting out).

Results.—Table 1 shows a general tendency for leaf length to increase from north to south but this trend is not consistent and populations from the same latitude may have very different mean leaf lengths.

As a first attempt to explain the geographical variation the populations were plotted on a map showing the distribution of average length of growing season (Figs. 1 and 2). It is apparent that there is a tendency for the longer leaved populations to occur in areas having a longer growing season. The map cannot, of course, show all variations in growing seasons especially that due to sudden changes in altitude, in fact, the correspondence with leaf length is better than this map suggests. A scatter diagram for the variation of leaf length with growing season is presented as Figure 3.

The correlation coefficients for each year taken separately and for both years taken together are given in Table 2. It seems reasonable to assume that leaf length is largely determined either by length of growing season or some other temperature controlled variable. As a matter of interest the correlation coefficients for leaf length on average annual temperature (A_2), average date of commencement of growing season (A_3) (days after 1st January), average May temperature (A_4), average accumulated growing season temperatures (A_5), are also given in Table 2. The partial regression coefficients for length (y) on A_2, A_4 and A_5 are given in Table 3 and of these three variables it would appear that the accumulated growing season temperature has the greatest influence on leaf length. There are, however, so many temperature measurements that could have been employed that it does not seem profitable to try to distinguish further between them. More interesting is the discovery of a possible reason for the greater leaf length in 1953 compared to 1950, which amounts to about 5 mm. for populations from sites having similar temperature régimes.

FIG. 1.—DISTRIBUTION OF LENGTH OF GROWING SEASON IN SCOTLAND (in days) (from Anderson and Fairbairn).

Open circles show sites of population collected in 1950.
Black ,, ,, ,, ,, 1943.

Numbers beside circles give mean leaf length in millimetres.

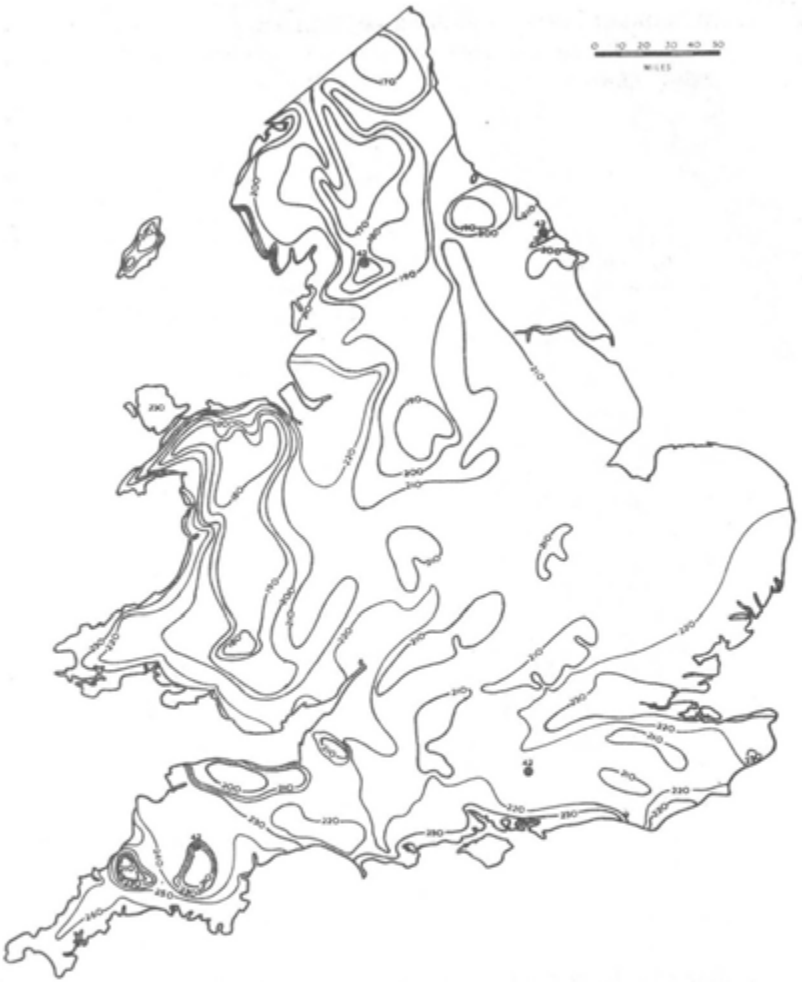

FIG. 2.—DISTRIBUTION OF LENGTH OF GROWING SEASON IN DAYS IN ENGLAND AND WALES.
(Explanation see Fig. 1.)

TABLE 2

TOTAL CORRELATION COEFFICIENTS BETWEEN VARIOUS TEMPERATURE MEASUREMENTS FOR AN AVERAGE YEAR AND LEAF LENGTH

	1950	1953	Both Years
A1 av. G.S.	+ .7276	+ .7537	+ .6053
A2 ann. temp.	+ .7665	+ .7625	+ .7145
A3 av. start	− .7109	− .7492	− .6113
A4 av. May	+ .6740	+ .7854	+ .6760
A5 av. accum.	+ .6813	+ .7675	+ .6525

TABLE 3

PARTIAL REGRESSION COEFFICIENTS FOR VARIOUS TEMPERATURE MEASUREMENTS FOR AN AVERAGE YEAR ON LEAF LENGTH

				t	p
A2	$b_{y\,2.45}$	+ 0.0396	± .03076	1.29	NS
A4	$b_{y\,4.25}$	+ 0.0756	± .03468	2.18	.05
A5	$b_{y\,5.24}$	+ 0.00324	± .0002823	11.47	.001

Considering first of all the effect of growing season, leaf length in any one year could conceivably be affected by the length of the growing season in the preceding year (Fig. 4) or by the date of commencement of the growing season in the year of collection (C_3) (Fig. 5). Comparison of Fig. 4 with Fig. 3 shows that with the present data the regression on length of preceding growing season does not serve to remove the difference between the two collections whereas that on C_3

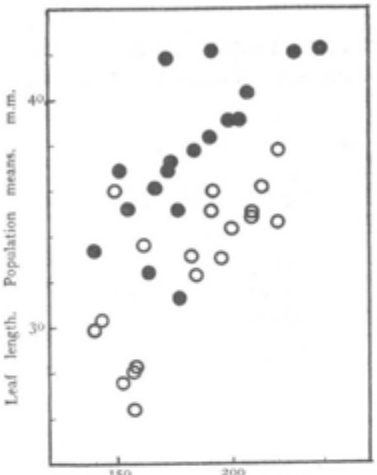

Fig 3

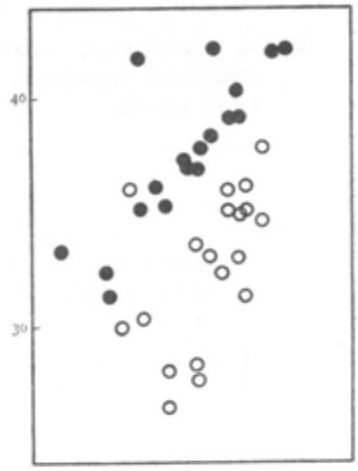

Fig 4

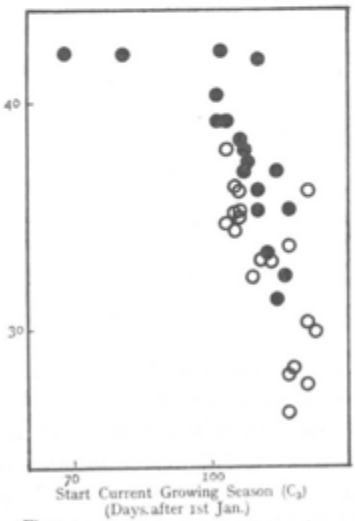

Fig 5

Figs 3-5 Relationship between leaf length and various aspects of Growing Season.
Open Circles 1950 Collection
Black Circles 1953 Collection

FIGS. 3, 4, AND 5.

removes a considerable portion of it and it seems probable that conditions in the current year (*i.e.* that in which the leaves were collected) are those most affecting leaf length in that year. If we now compare for the current year, date of start of growing season (C_3), mean May temperature (C_4) and accumulated growing season temperature (C_5), each with the corresponding value for an average year A_3, A_4 and A_5, we can calculate the three pairs of partial regression coefficients in Table 4. In each case it will be seen that it is the coefficient for the current year that shows the greater effect on leaf length, the partial regression coefficients for the current year all reaching the 1 per cent level of significance while those for an average year are all non-significant and are, in fact, for all three variables less than their standard errors.

TABLE 4

PARTIAL REGRESSION COEFFICIENTS FOR AVERAGE AND CURRENT TEMPERATURE MEASUREMENTS ON LEAF LENGTH

	Average year			Current year		
	$b_{y_{a.c.}}$	t		$b_{y_{c.a.}}$	t	
3	+ .04340 ± .0787		NS	− .336303 ± .100481	3.35	<.005
4	− .09193 ± .3134		NS	+ 1.50383 ± .2578	5.83	<.001
5	− .000920 ± .0017		NS	+ .005949 ± .00216	2.75	<.01

In the light of the foregoing it seems apparent that temperature and leaf length in *B. pubescens* are highly correlated and since by multiple regression it is possible to account for approximately 75 per cent of the sum of squares due to variation in population mean leaf length, the relationship is probably one of cause and effect. It also seems probable, though this is less certain, that year to year variations in leaf length are due to parallel variations in temperature during the early growing season and do not seem to be much affected by temperature conditions of the preceding growing season. Data for more

than two years would be necessary before any firm conclusions could be reached on this point.

The Garden populations.—The seven garden populations were sown in boxes in the late autumn of 1953 and were planted in the garden in March 1955. The first measurements of leaf length were made in 1959. The parent populations came from a fairly wide range of temperature regions within Scotland and one came from one of the mildest parts of England (Wareham, South Dorset). The differences between populations are significant at the 1 per cent level but there is no significant correlation between leaf length and the length of growing season of the location of the parent populations.

The high correlation which exists in the wild disappears under the uniform conditions of the experimental garden. This result, plus the finding reported above, of the relatively great importance of conditions in the current season in determining leaf length, would indicate that the portion of the variation in length due to temperature variation is only to a small extent under genotypic control.

Summary and Conclusions

(1) A correlation between variation in leaf length in *B. pubescens* and growing season temperature has been demonstrated and it is considered likely that some function of temperature largely controls leaf length.

(2) It is likely that the portion of variation calculated with temperature is not under genotypic control since leaf length varies from year to year in step with temperature and different genotypes grown in a uniform environment do not show any significant correlation with the temperature régime of their original habitat.

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A SIMPLE METHOD FOR THE SAMPLING OF NATURAL POPULATIONS

JENS CLAUSEN

Department of Plant Biology, Carnegie Institution of Washington, Stanford, California

Recent papers in this report and elsewhere (Harberd, 1957, 1958; Wilkins, 1959) have appropriately raised a discussion about the adequateness of sampling techniques in genecology. Sampling methods vary with the investigators, and the results may be influenced by the sampling technique. Since there is no formal record of the sampling methods applied by our group, the principles of these methods will be stated in the following pages.

At the beginning of our investigations some 25 years ago the merits of various procedures were considered, and it was decided that the most representative and simplest sampling method would be by mass collection of seed from a great number of individuals evenly scattered through the population. Such a method is applicable to annuals and perennials, and to self-pollinators, cross-pollinators, and seed apomicts.

Geneticists have generally been trained to test the progeny of individual plants rather than to sample the variability of the population as a whole. Executing sampling by progeny testing of individual plants is, however, a laborious method, greatly reducing both the number of individuals and the number of populations that can be sampled. Sampling by individual plants is liable to produce erratic results that do not properly represent the population.

It should be emphasized that a sampling of the variability within a population is not to be confused with a test of the breeding structure of the population. Investigations on the breeding structure require progeny tests of individual plants after open pollination, self-pollination, or mutual pollination, and are best conducted as step number two in the analysis.

The early transplant investigators, such as Göte Turesson and Harvey M. Hall, used direct transplants from the wild. Turesson's samples were 20 individuals per population. These investigators used the direct transplantation method, because

they wanted to study the performance of plants that actually had grown in the population, representing the immediate results of natural selection.

It can be objected, however, that in any one year the individuals that grow in a habitat compose only a part of the potential population that occupies that habitat over a period of years. In any one habitat the climate and other environmental factors vary somewhat from year to year. The biotype composition of a habitat has adjusted itself to the potential fluctuations within the habitat and varies accordingly. This fluctuating variability is especially noticeable in habitats populated by annuals. A properly drawn seedling sample is therefore a truer over-all representation of the biotypes that may occupy the habitat over a period of years than an equally large sample of direct transplant individuals taken in a single year.

A sample of 20 plants from a population is generally too small and does not provide an adequate impression of the variation within the population. In most species, similarly to Gregor *et al.* (1936), we have found that somewhere between 60 and 100 plants per population is a minimum sample. Duplicates among the phenotypes are rare within samples of that size.

Seed samples are best collected at a time when the largest number of plants have ripe seed. Seeds are harvested as far as possible over the entire population, except when it is miles across. In sampling, one walks forth and back through the population in a zig-zag pattern, gathering handfuls of seed material at even intervals. Generally, the aim is to sample the effective breeding unit. Separate samples of fractions of a population have sometimes been taken when uneven terrain or other unequalities in the environment suggest possible differences within the population. Such fractions, however, have seldom proved to be statistically distinguishable in the experiment garden. It is generally more useful to sample a larger number of populations than to conduct highly detailed analyses of a few populations.

At the laboratory each sack of seed material from a population is cleaned by passing the collected material through screens and by blowing the chaff away. The sacks of material accordingly become reduced to convenient bags of cleaned seed, each representing a natural population. The cleaning

process randomizes the seed from various sectors of the population.

Samples of the seeds are sown in seed pans, the aim usually being to obtain about 300 to 600 seedlings per population. About 120 seedlings are normally pricked into small flats where they are spaced evenly. Selection during the pricking process is carefully avoided by systematically planting the seedlings from one side of the seed pan until the desired number of plants is obtained. The remainder are kept as a reserve. Selection is similarly avoided at the time when the seedlings are transferred from the flats to the garden rows. The plants in the experiment garden represent therefore fair samples of the potentials within the natural populations.

A sound sampling is considered more important than using refined statistical techniques on poorly sampled material. It is also important to look for recombinations of minute individual differences within a population, suggesting subtle genotypic variabilities that may escape the traditional statistical analysis. Statistical methods are at best only crude tools for the determination of phenotypic variability. The variation curve is a better biometrical description of the variability of a character within a population than are statistical means, variance, and standard deviations that tend to submerge the pattern of the variability. Progeny analysis and cytological methods provide powerful tools that supplement the descriptive methods.

Harberd (1957) emphasized that individuals of plants that spread by rhizomes, such as *Festuca rubra* and *Poa pratensis*, may occasionally occupy a large sector of a population. A biologist should be aware of such a situation and use common sense in adjusting his sampling method to the species. As an example, when Wm. M. Hiesey in 1943 investigated, by the clonal method, a population of the highly rhizomatous *Poa pratensis* in a large meadow near the Mather transplant station, he sampled at wide distances with the result that all the 12 plants he collected belonged to distinct individuals. They differed in colour of leaves, in susceptibility to rust and mildew, and in their date of flowering. In chromosome numbers they ranged between 68 and 50, and they varied also greatly in their degrees of apomixis, one plant being completely sexual.

Populations of self-pollinating plants have likewise been found to be composed of a fair number of biotypes. Years ago

TABLE 1

Madia sativa, A SELF-POLLINATING POPULATION FROM NEAR FREESTONE, SONOMA COUNTY, CALIFORNIA.

Variation in stem height in the original culture and in three selfed progenies when grown in a uniform garden at Stanford, California.

From original seed		Progenies from selfing		
Height, cm.	2670 ex Freestone	3142 ex −2, 134 cm.	3143 ex −3, 102 cm.	3144 ex −4, 76 cm.
160				
150	.	1	.	.
140	8	20	.	.
130	13 −2	33	.	.
120	14	18	.	.
110	22	10	2	.
100	19 −3	1	24	.
90	17	3	42	.
80	5	1	15	2
70	2 −4	.	9	20
60	.	.	6	52
50	.	.	.	16
40	.	.	1	7
30	.	.	1	3
Total	100 Variable	87 Early	100 Late	100 Medium late
		Each highly uniform		

it was shown that wild populations of *Viola arvensis* and of the *Erophila verna* complex contained several distinct biotypes. The distinct chromosome numbers of the *Erophila* forms provide irrefutable evidence of this fact (Winge, 1940).

Similar intra-population variability was observed and documented in the self-pollinating subspecies *typica* and *reflexa* of *Potentilla glandulosa* (Clausen and Hiesey, 1958). In the California Madiinae of the sunflower family, several species of *Layia* and *Madia* are habitually self-pollinated although the majority of the species of these genera are self-incompatible.

The populations of the self-pollinating species *Layia hieracioides* and *L. paniculata* and those of the *Madia sativa* complex are commonly composed of several, and possibly many, distinct biotypes. Contradicting what has been postulated, the self-fertile species are among the most dependable and constantly occurring of the Madiinae, whereas the erratically occurring and rare species happen to be self-incompatible.

As an example of a population study by the seed sample and progeny method, Table 1 presents an analysis of a population of *Madia sativa*, the culture 2670 from near Freestone, Sonoma County, California. Only the character of stem height was used in the statistical description, but the plants grown from the seed collected in the wild differed also in their time of flowering, in the degree of the divergence of the branching, and in their degree of pilosity. Three plants of 2670, —2 to —4, were enclosed in garden cages to protect them against rare cross-pollinations. Their progenies, 3141 to 3144, were grown the following year with the results shown in the Table. It was observed that the three progenies were highly distinct from each other, whereas the plants within each progeny were highly uniform, and much more so than the plants grown from the original seed. The actual number of biotypes in the Freestone population is not known, except that it is three or more. On account of the many seeds produced by each homozygous individual it seems likely that the sample of 100 plants in 2670 was too small to include the rare biotypes.

Why are there so many biotypes in populations of apomictic and self-pollinating species, although these species are capable of multiplying a single, highly successful biotype? Possibly because populations that are rich in biotypes have a long-range selective advantage over others that possess only few. Distinct biotypes mature at different times, or they differ slightly in their demands of soil and climate, some being at an advantage in certain years and others in others. A population evolves as the result of selections over many years and not on the basis of a single year's success.

Absolutely obligate apomicts and self-pollinators probably do not exist. Measured on the evolutionary time scale, a crossing every ten, hundred, or thousand years can produce considerable variability.

Cross-fertilization is not even excluded after self-pollination has occurred. A simple experiment performed in 1920 on the

Line 637 of *Viola arvensis* illustrates this point. In *arvensis*, self-pollination occurs before the flowers have opened. This particular *arvensis* line was established from a distinctive individual in the variable population C from Tysinge Moor, Sjælland, Denmark (Table 1 in Clausen, 1921). Five already opened flowers on a plant of this pure line were also pollinated by pollen from a plant of *Viola tricolor* from the population H taken in a spruce plantation in Sophienholm Hills a few miles away. Three of the *arvensis* flowers were cross-pollinated on the first morning after they had opened and produced 36 hybrids and 54 non-hybrids; a two-day-old flower resulted in 3 hybrids and 30 non-hybrids; the fifth flower was probably three days past anthesis and produced selfed *arvensis* only. This experiment suggests that in its growth through the *arvensis* pistil, the *tricolor* pollen can outgrow that of *arvensis* itself, thereby producing hybrids that contribute to the variation of both species in the wild.

In the *Madia sativa* complex (the 16-chromosome *M. sativa*? *capitata* and *gracilis*) the stigmas are usually powdered by pollen before the flower heads open. It was not uncommon, however, to find 1-3 per cent hybrids among plants developed from seeds of unprotected flower heads, irrespective of whether the seeds had been harvested in the natural habitats or in the experiment garden. Some of the hybrids were inter-specific although partly fertile. Genes from all three species can therefore constantly add to the variability in these otherwise self-fertilising plants.

Conclusions.—A method of sampling wild populations by mass collection of seeds and growing generous samples of plants from them is described, and its advantage over the direct transplant method is discussed. A more thorough sampling is afforded by the seedling method than by direct transplantation only, revealing multitudes of biotypes in populations of cross-pollinating species, and fair numbers of distinct biotypes in populations of habitually apomictic and habitually self-pollinating species. It is emphasized that completely obligate apomicts and self-pollinators probably do not exist. Data on *Madia, Layia, Viola, Erophila, Potentilla* and *Poa* are discussed.

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INTERNAL BREEDING BARRIERS IN *PAPAVER*

I. H. McNaughton

Papaver species are frequently found in Britain as weeds of arable land and waste places, especially on the lighter, more calcareous soils. All the species have probably been introduced and appear to have spread northwards into Britain from their centre of origin in the Eastern Mediterranean. Poppies are particularly associated with the growing of cereal crops but are becoming rarer due to the advent of selective weed-killers.

Six species, five British and one continental, were selected for study as showing a fairly close systematic relationship. All belong to the *Rhoeades* as classified by Fedde (1909) and may be subdivided on characters of the capsule.

Orthorhoeades—smooth capsuled

1. *P. rhoeas* L. . capsule almost globose.
2. *P. dubium* L. . capsule elongate ; sap white.
3. *P. lecoqii* Lam. . capsule elongate ; sap yellow.

Argemonorhoeades—bristly capsuled

4. *P. argemone* L. . capsule elongate, bristles few.
5. *P. hybridum* L. . capsule globose, bristles many, long and spreading ; petals narrow, deep red ; pollen pale blue.
6. *P. apulum* Ten. capsule globose, bristles many, short and erect ; petals broad, orange-pink ; pollen blue-black.

Clear-cut morphological distinctions are difficult to make between *P. dubium* and *P. lecoqii*. *P. lecoqii* has frequently been regarded as a variety of *P. dubium*. The differentiation of these two species is discussed in detail elsewhere (McNaughton and Harper, 1960c).

It is not unusual to find several species growing in the same habitat. *P. rhoeas* and *P. dubium* sometimes occur together in considerable numbers amongst cereals and on the sandier soils *P. argemone* may be associated with them. On occasions four or five species may be found within a few feet of each other on waste ground. All the species are annuals and flower over the same period, all are entomophilous and have insect visitors in common (McNaughton and Harper, 1960a).

Inter-specific hybridisation might reasonably be expected to occur, but only one authentic record of a natural *Papaver* hybrid, a specimen attributed to a cross between *P. rhoeas* and *P. dubium*, has been reported (Salmon, 1919). An examination of a large number of habitats in which two or more *Papaver* species were present showed that hybrids are indeed extremely rare in the field (McNaughton and Harper, 1960a).

The fact that natural hybrids are seldom found, yet some can readily be produced artificially (see below), suggests that there are factors operating which reduce the chances of inter-specific pollinations. These external barriers to hybridisation have been shown to include a high degree of self-pollination in some of the species and the discriminatory behaviour of the principal insect pollinator, the honey-bee (McNaughton and Harper, 1960a).

Internal barriers, occurring after inter-specific pollinations have taken place, and acting either in preventing the formation of hybrids or in reducing their viability or fertility, may best be studied by making artificial crosses under controlled conditions. The plants used in these experiments were all grown from seed collected in the field, with the exception of *P. apulum* (a continental species) which was obtained from the Botanic Garden, Cambridge. Seed of *P. rhoeas*, *P. dubium*, *P. lecoqii* and *P. argemone* was collected from wild populations in the Oxford area. Seed of *P. hybridum* was taken from plants near Cambridge.

Plants were raised to maturity in a cool greenhouse and artificial crosses effected by emasculating buds and applying pollen of the appropriate male parent by means of a fine brush. Pollen was brushed over each of the stigmatic rays in order to facilitate maximum seed set. Glassine bags were used to exclude all other pollen. Since conditions within the bags might have an adverse effect on the success of any cross a series of cross-pollinations within each species were carried out using the same technique. In each of these intra-species crosses high seed set was obtained. It was therefore concluded that failure of an inter-species cross was more likely to be due to genetic differences between the parents than to physical factors of the environment. Results are shown in Table I and the crossing relationships of the species are represented diagrammatically in Fig. 2. An estimate of the degree of stimulation of ovary and seed development was obtained for each cross

TABLE I

RESULTS OF INTRA- AND INTER-SPECIFIC CROSSES IN *PAPAVER*

Cross	No. of attempts	No. of successful attempts	% Success	Degree of stimulation	Mean No. of seeds per capsule*
P. rhoeas × *P. rhoeas*	7	7	100.0	5	410.9
P. rhoeas × *P. dubium*	67	29	43.2	5	52.4
P. rhoeas × *P. lecoqii*	37	11	29.7	4	36.3
P. rhoeas × *P. argemone*	10	0	nil	1	..
P. rhoeas × *P. hybridum*	8	0	nil	1	..
P. rhoeas × *P. apulum*	36	0	nil	1	..
P. dubium × *P. dubium*	4	4	100.0	5	829.3
P. dubium × *P. rhoeas*	35	31	88.6	5	360.1
P. dubium × *P. lecoqii*	37	33	89.2	5	460.4
P. dubium × *P. argemone*	15	5	33.3	3	30.8
P. dubium × *P. hybridum*	4	4	100.0	3	1.3
P. dubium × *P. apulum*	25	13	52.0	3	26.4
P. lecoqii × *P. lecoqii*	8	8	100.0	5	1284.8
P. lecoqii × *P. rhoeas*	37	27	73.0	4	19.8
P. lecoqii × *P. dubium*	51	49	96.1	5	698.4
P. lecoqii × *P. argemone*	6	0	nil	1	..
P. lecoqii × *P. hybridum*	5	0	nil	1	..
P. lecoqii × *P. apulum*	10	0	nil	1	..
P. argemone × *P. argemone*	6	6	100.0	5	211.8
P. argemone × *P. rhoeas*	22	0	nil	0	..
P. argemone × *P. dubium*	33	0	nil	1	..
P. argemone × *P. lecoqii*	12	0	nil	0	..
P. argemone × *P. hybridum*	15	0	nil	1	..
P. argemone × *P. apulum*	55	33	60.0	3	60.0
P. hybridum × *P. hybridum*	5	5	100.0	5	493.0
P. hybridum × *P. rhoeas*	8	0	nil	0	..
P. hybridum × *P. dubium*	5	0	nil	0	..
P. hybridum × *P. lecoqii*	8	0	nil	0	..
P. hybridum × *P. argemone*	18	0	nil	0	..
P. hybridum × *P. apulum*	37	0	nil	0	..
P. apulum × *P. apulum*	8	8	100.0	5	98.0
P. apulum × *P. rhoeas*	111	0	nil	0	..
P. apulum × *P. dubium*	95	0	nil	0	..
P. apulum × *P. lecoqii*	17	0	nil	0	..
P. apulum × *P. hybridum*	15	0	nil	0	..
P. apulum × *P. argemone*	51	3	6.0	3	1.0

* In successful attempts

(see Table I) and scored as follows : (0) capsule not stimulated, shrivelling before maturity, no seed set ; (1) capsule swollen, seeds represented by fine dust ; (2) capsule swollen, seeds flattened, lacking in content and apparently non-viable ; (3) capsule swollen, a few apparently viable seeds ; (4) capsule swollen, some apparently viable seeds, many others flattened and lacking in content ; (5) capsule swollen, seeds abundantly produced and all apparently viable. (0) and (1) indicate failure of pollen to germinate or, if germinated, the inability of the pollen tube to travel down the style and fertilize the egg cell. (2), (3) and (4) suggest varying degrees of embryo abortion or endosperm deficiency following fertilization, whilst (5) is the normal state to be expected in intra-species crosses.

The following F_1 hybrids were raised to maturity : *P. dubium* × *P. rhoeas*, *P. rhoeas* × *P. dubium*, *P. dubium* × *P. lecoqii*, *P. lecoqii* × *P. dubium*, *P. lecoqii* × *P. rhoeas* and *P. argemone* × *P. apulum*. It is noteworthy that hybrids were only produced between species belonging to the same subsection of the genus. Crosses within the *Orthorhoeades* were particularly successful, the only inter-specific combination which failed to produce a hybrid was *P. rhoeas* × *P. lecoqii*. In the *Argemonorhoeades* only the cross between *P. argemone* and *P. apulum* yielded a hybrid. All crosses between species of the *Orthorhoeades* and the *Argemonorhoeades* failed to produce hybrids, thus the two groups were shown to be effectively isolated.

Examination of pollen and attempted back-crosses to the parents revealed that the hybrids *P. lecoqii* × *P. dubium* and *P. dubium* × *P. lecoqii* were less than 5 per cent fertile (McNaughton and Harper, 1960c), *P. argemone* × *P. apulum* plants were entirely pollen sterile but produced a few seeds when parental pollen was applied to hybrid stigmas. The single individual raised from the cross *P. lecoqii* × *P. rhoeas* was completely sterile. *P. dubium* × *P. rhoeas* and *P. rhoeas* × *P. dubium* hybrids were not only 100 per cent infertile but many of the plants were inviable (McNaughton and Harper, 1960b).

Since differences in chromosome number are frequently effective barriers to gene flow between sympatric species, a cytological examination of *Papaver* species and artificial hybrids was carried out. With the exception of *P. rhoeas* (Lawrence, 1930) no counts had been made on British material. It was

also necessary to ascertain the position of *P. lecoqii* which had often been confused with *P. dubium*.

Small, unopened buds were selected, the outer coverings dissected off and the anthers fixed in Carnoy's solution, the material was then stored in a deep freeze incubator at −10°C for two to three months. Individual anthers were squashed in aceto-carmine solution with ferric chloride added as a mordant. Chromosomes of each of the species at meiotic metaphase I are shown in Fig. 2. The following counts were obtained :—

Orthorhoeades		*Argemonorhoeades*	
P. rhoeas	2n = 14	*P. apulum*	2n = 12
P. lecoqii	2n = 28	*P. hybridum*	2n = 14
P. dubium	2n = 42	*P. argemone*	2n = 42

In the hybrid *P. dubium* × *P. rhoeas* counts of 2n = 28 (21 + 7) were made whilst in *P. argemone* × *P. apulum* counts of 2n = 27 (21 + 6) were obtained. 35 chromosomes (14 + 21) were found in the hybrid *P. lecoqii* × *P. dubium*. *P. dubium* × *P. rhoeas* plants showed trivalents and bivalents on the metaphase plate, together with unpaired univalents scattered around the periphery of the cell. Three cells were scored as follows : (1) I_{III}, 7_{II}, II_{I}; (2) I_{III}, 7_{II}, II_{I}; (3) 3_{III}, 4_{III}, II_{I}. At later stages of division lagging chromosomes were seen to be excluded as micro-nuclei. In *P. argemone* × *P. apulum* hybrids an average of six bivalents per cell was observed at metaphase I with a number of unpaired chromosomes scattered around them. At anaphase and telophase several univalents were seen to lag behind and divide later than the bivalents. Other univalents did not undergo disjunction and travelled undivided to the poles where they appeared to be included in the daughter nuclei. In some cases a few univalents remained behind in the centre of the cell. The anomalous meiotic behaviour of the chromosomes in the hybrids studied would adequately account for their lack of pollen fertility.

Within the *Orthorhoeades* there is a simple polyploid series : *P. rhoeas* (diploid), *P. lecoqii* (tetraploid) and *P. dubium* (hexaploid), *P. lecoqii* and *P. dubium* are clearly differentiated as separate species. In the *Argemonorhoeades* *P. apulum* is peculiar in possessing only 12 chromosomes, the basic number for the group is considered by Darlington and Wylie (1945)

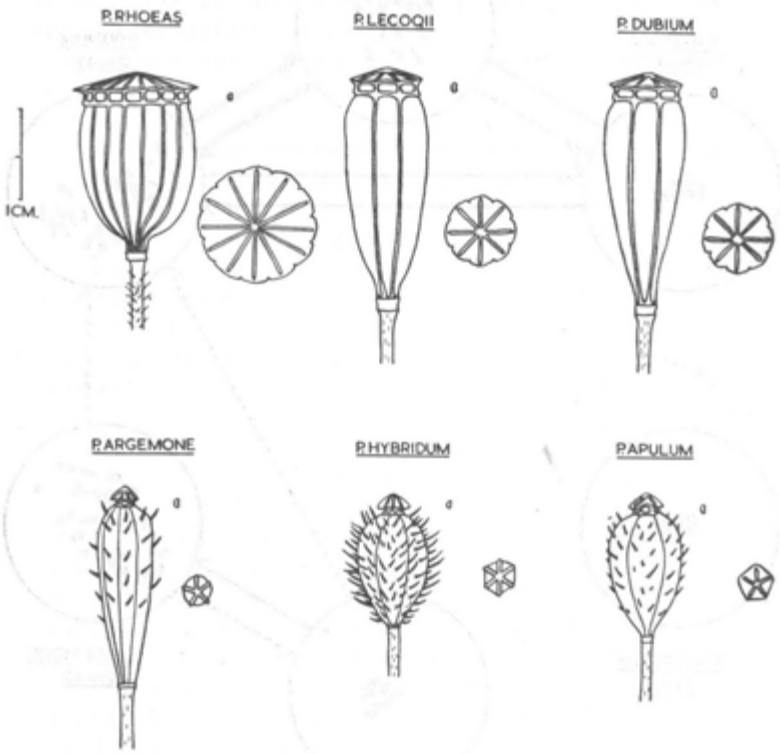

FIG. 1.—CAPSULES OF *PAPAVER* SPECIES.
Stigmatic discs are shown at right of each capsule.

F

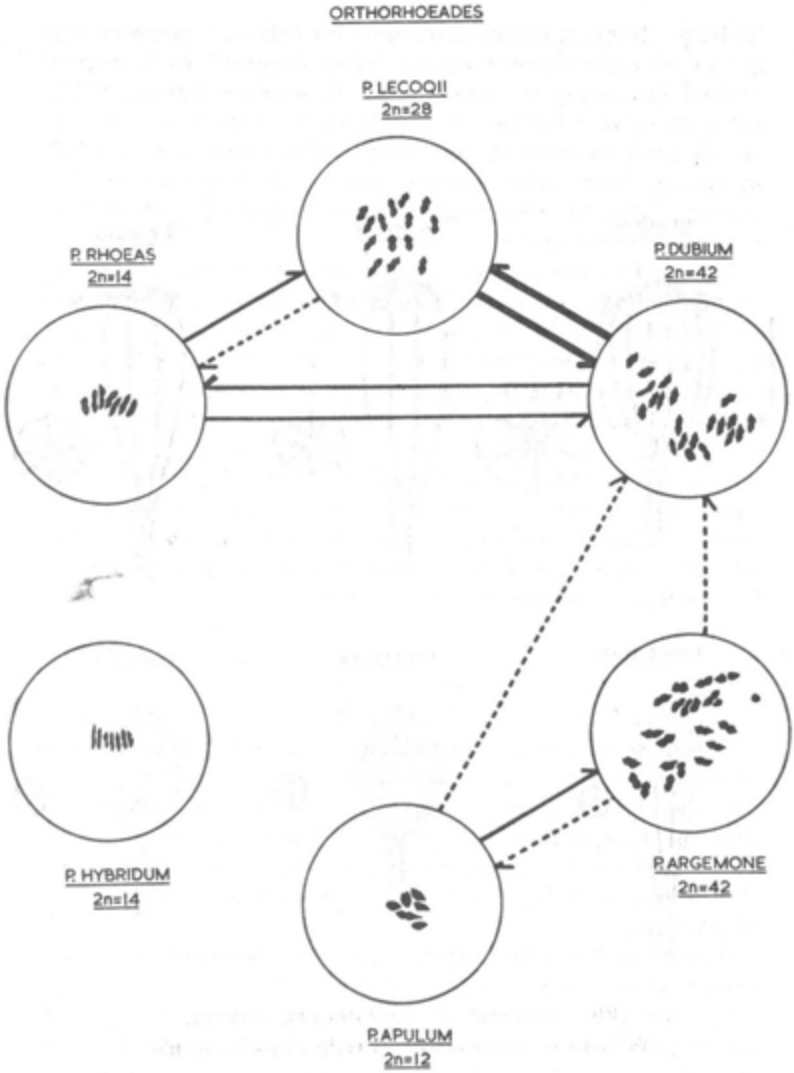

FIG. 2.—CROSSING RELATIONSHIPS OF *PAPAVER* SPECIES.

Thick line—F_1 hybrids partially fertile.
Thin line—F_1 hybrids sterile.
Dotted line—seed produced but failed to germinate.
Arrows indicate direction of crosses.
Chromosomes are reproduced × 700 approx.

to be 7. Sugiura (1940) comments on this and suggests that fusion of chromosomes at one time occurred in a normal diploid (2n = 14) to give rise to *P. apulum* (2n = 12).The presence of one bivalent larger than the others (see Fig. 2) would tend to support this view. The chromosomes of *P. hybridum* appear to be somewhat smaller than any of the other species. This has also been noted by Sugiura (1940) working with continental material.

External barriers preventing or reducing inter-specific pollinations in *Papaver* are likely to operate under field conditions but should these break down internal barriers due to genetic differences between the species would tend to prevent hybrids being formed in certain inter-specific combinations. In others, although hybridisation is possible, chromosomal differences would result in a high degree of sterility so that the chance of gene flow between the species would be almost negligible. Although *Papaver* species are frequently found together in the same habitat, flower over the same period and have insect visitors in common, hybridisation is considered unlikely and the possibility of introgression remote.

Summary

Chromosome numbers were recorded for six *Papaver* species as follows: *Orthorhoeades* (smooth-capsuled group)—*P. rhoeas* 2n = 14; *P. lecoqii* 2n = 28; *P. dubium* 2n = 42. *Argemonorhoeades* (bristly-capsuled group)—*P. apulum* 2n = 12; *P. hybridum* 2n = 14; *P. argemone* 2n = 42. All material, except *P. apulum*, was derived from wild populations in the south of England.

Every possible inter-specific cross was attempted and results expressed as mean numbers of seeds obtained per capsule. The crosses *P. rhoeas* × *P. dubium* (and reciprocal) and *P. rhoeas* × *P. lecoqii* gave rise to completely sterile hybrids. *P. dubium* × *P. lecoqii* (and reciprocal) and *P. argemone* × *P. apulum* plants were less than 5 per cent fertile. All other inter-specific combinations failed to produce hybrids. Intra-specific crosses carried out under the same conditions gave high seed yields. The meiotic behaviour of some of the hybrids was studied. Results are discussed with regard to the genetic isolation of the species in nature.

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THE MEASUREMENT AND GENETICAL ANALYSIS OF LEAD TOLERANCE IN *FESTUCA OVINA*

D. A. Wilkins

1. Introduction

For the past four years work on the genecology of lead tolerance in *Festuca ovina* has been carried out at this station. This paper is intended not to present detailed conclusions but to summarise the results so far obtained, and to suggest possible interpretations. In spite of the amount of effort devoted to refining the measurement of tolerance the nature of the genetic mechanism controlling it has not been established with certainty. It will be suggested that on balance the evidence points to a major gene system, but the clear discontinuities which would be needed to analyse this exactly have not so far been found. Some of the wider genecological aspects have been discussed elsewhere (Wilkins, 1960).

2. Material

Vegetative samples of *F. ovina* were collected from several populations in each of the chief lead mining areas of Britain, together with soil samples, and the analysis of the latter showed the presence of varying amounts of heavy metals, with lead usually predominating. The original plants have been preserved in the greenhouse since collection by regular tillering and transplanting, and each year a large number of crosses has been made. As soon as it became clear that tolerance tended to be dominant over non-tolerance it was possible to concentrate on crossing a selection from the previous year's progenies with non-tolerant plants.

Unfortunately, the tolerant diploid was discovered a year later than the tetraploid, and so the results to be discussed are derived from two generations of crossing with the tetraploid but only one with the diploid. Owing to the moderate self-sterility of the species selfed progenies were difficult to produce, but it had the advantage that it was not necessary to emasculate for crossing, and the panicles from two plants could simply be enclosed together in a bag just before the flowers opened.

The actual degree of self-fertility was determined by enclosing a group of panicles from each plant in a separate bag and sowing the resulting seed. A number of self-fertile plants were found and these had to be discarded as female parents for crossing. A few of them were so highly self-fertile that their selfed progenies were themselves big enough to be raised and tested for tolerance.

3. Methods of testing

Root elongation was chosen as a sensitive indicator of poisoning by metals in the soil, and the results of some of the preliminary experiments with lead in culture solutions have already been reported (Wilkins, 1957). They may be briefly summarised here.

The addition of calcium was found to reduce the toxicity of a solution of lead nitrate by a factor of more than 20, in that while 1 ppm. of lead by itself was enough to stop root growth a tolerant plant could still grow at 25 ppm. in the presence of calcium. The mechanism of this buffering remains unknown, but its existence greatly simplified all the subsequent experiments because it overcame the unexpected difficulty of maintaining the concentration of solutions containing only 1 ppm. of lead. The presence of such organic tissues as roots, tillers and contaminant algae led to a rapid removal of lead from these solutions, so that after a few hours the concentration was too low to be measured by the usual method of sulphide colorimetry. The higher concentration which was needed in the presence of calcium to produce a given effect on the plants did not decline by more than about 5 ppm. over a period of several days. The 4.24 molar solution obtained by adding 1 g. hydrated calcium nitrate to the litre was found to be close to the optimum, and was used for all the experiments and tests reported in this paper.

Two possible ways of measuring lead tolerance were considered. If each plant had been tested in a series of lead concentrations some critical value above which growth was impossible might have been found. Apart from the obvious difficulty of finding a suitable end-point in such a test, and the labour of keeping a large number of solutions accurately standardised, the danger of dividing up a continuous range of

variation into a series of arbitrary steps made any such "threshold" method unacceptable. Instead of using a series of test solutions the alternative method of comparing the rates of growth in a single concentration of lead was adopted. For a continuous scale to be reliable it was necessary to ensure that growth in the most tolerant plant was slightly slowed down while that in the least tolerant was not stopped altogether, and a concentration of 25 ppm. of lead and a test period of four days were chosen on this basis. In order to allow for differences between absolute growth rates in the absence of lead an Index of Tolerance was constructed from the measurements of two growth increments on the same root—one over two days in calcium nitrate, and one over four days after the addition of lead. The markedly skewed distribution of the ratio of these growth rates was made symmetrical by converting to logs. The Index was thus calculated as follows :—

$$I = 1 + \log \frac{2B}{D}$$

B = growth in 2 days in calcium nitrate.
D = growth in 4 days in calcium nitrate + 25 ppm. Pb. (as nitrate).

This Index had high values around 2.0 (representing a reduction of growth rate to 10 per cent of normal) for non-tolerant plants, and low values around 1.2 (a reduction to 63 per cent) for highly tolerant plants.

Tillers from plants in the greenhouse had their roots removed and were put into 9-inch lengths of glass tubing open at both ends. The tubes were placed upright in a culture solution until new roots had grown, and were then passed through the two test solutions. At each transfer the length of the longest root in each tube was measured, so that all references to root growth are to growth in length down the tube, and throughout the experiments the temperature in the controlled environment chamber was kept at 21 degrees C. and the light from the bank of fluorescent tubes at 400 f.c. intensity. The only direct aeration to the roots was that provided during the transfers to fresh solutions—usually at intervals of two days.

Towards the end of the experiments to be reported here it was found that the concentration of lead inside the tubes sometimes fell to a much lower value than that of the surrounding liquid in the course of one or two days. This is now being overcome, and the aeration improved, by a device for raising

and lowering the tubes at frequent intervals, but this is only in the experimental stage and does not affect any of the results under discussion. In spite of the poor aeration inside the undisturbed tubes it was found that most roots grew steadily for at least a week as long as the tiller itself remained healthy. Linear growth rates of 10 mm. in 24 hours were quite usual in the calcium nitrate solution, and the average throughout the tests would be perhaps 5 mm. per day.

The Index defined above was calculated for the longest root in each tube. If two or more tubes per plant had been set up the values for each plant were then averaged, and in many cases an analysis of variance was carried out with the individual indices.

4. Results of testing wild plants

It was clear from the start that the early hopes of discovering either a clear division into tolerant and non-tolerant, or a completely continuous range from one to the other, were not to be easily fulfilled. Every plant available was first tested once only and the means and standard deviations of the populations calculated, and the large size of the latter reflected a big and seemingly continuous range within each population. It was important to decide whether this range was a genetical one or merely the reflection of a high experimental error, but the amount of repetition of the test which has so far been possible with these original plants has not provided a conclusive answer. The different populations from Leadhills, for example, differed clearly in their means, but the differences between their constituent plants were only significant at the 5 per cent level and so can hardly be said to be conclusively established.

Amongst the Pennine populations an interesting relationship was found between the means and standard deviations (Fig. 1). The samples were too small (6-12 plants each) for the values to be known with precision, but the population means tended to be crowded towards the two ends of the scale of tolerance. Those with intermediate means were not only few in number but they had the highest standard deviations. This pattern is most easily explained if it is assumed that the extreme populations were each made up of one type of plant only,

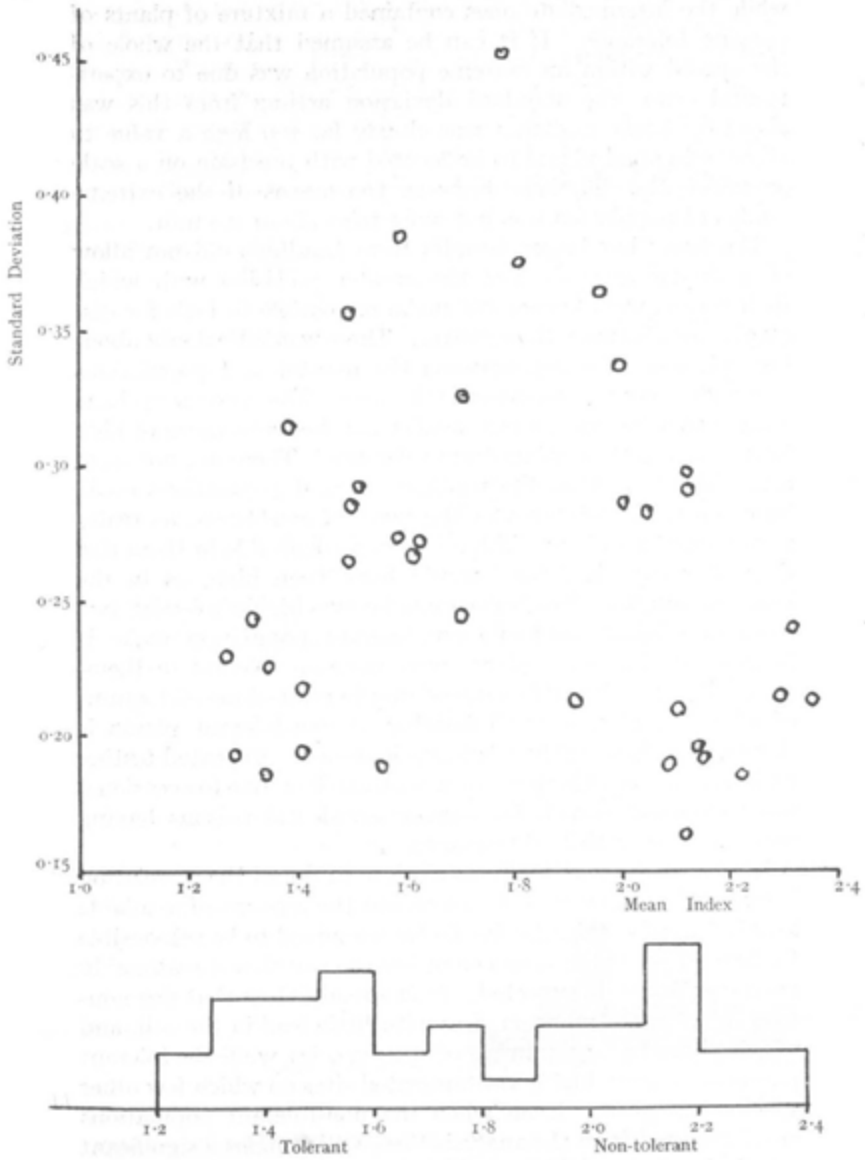

FIG. 1.—DISTRIBUTION OF MEAN TOLERANCE INDICES OF PENNINE POPULATIONS, AND RELATIONSHIP WITH STANDARD DEVIATION. (6-12 plants each.)

while the intermediate ones contained a mixture of plants of varying tolerance. If it can be assumed that the whole of the spread within an extreme population was due to experimental error, the standard deviation arising from this was about 0.2 units, and that was clearly far too high a value to allow individual plants to be located with precision on a scale on which the difference between the means of the extreme groups of population was not more than about 1.0 unit.

The fewer but larger samples from Leadhills did not allow of a similar analysis, but the greater precision with which their means were known did make it possible to look for discontinuities between those means. There was little doubt about the existence of a gap between the non-tolerant populations and those showing moderate tolerance. The more surprising feature was the similar but smaller gap between those of high tolerance and those of medium tolerance. There did not seem much likelihood that the medium tolerant populations could be made up of mixtures of the two extreme types, as really non-tolerant plants would have been recognisable in them and their standard deviations would have been high, as in the Pennine samples. The histograms for two highly tolerant, two medium tolerant, and one non-tolerant population make it fairly clear that some plants were medium tolerant in themselves (Fig. 2). It will be noticed that in most of the histograms of Figs. 2 and 3 a small number of non-tolerant plants is shown, even in a highly tolerant population. Repeated testing made it fairly certain that this was an artefact, due to occasional roots stopping growth for various accidental reasons having no connection with lead tolerance.

It is intended eventually to analyse in detail the correlation between the tolerance of *F. ovina* and the amount of available lead in the soil—the selective factor presumed to be responsible for tolerance—but it is sufficient here to say that a reasonably good correlation is expected. It is already clear that the non-tolerant plants come from sites with little lead in the soil, and which support a large number of other species, while the tolerant plants come from highly contaminated sites on which few other species can grow. Even when the non-tolerant populations are eliminated from the analysis there still remains a significant correlation between the mean tolerance of a population and the amount of lead in the soil. This affords support for the idea that there is more than one type of tolerant plant. For

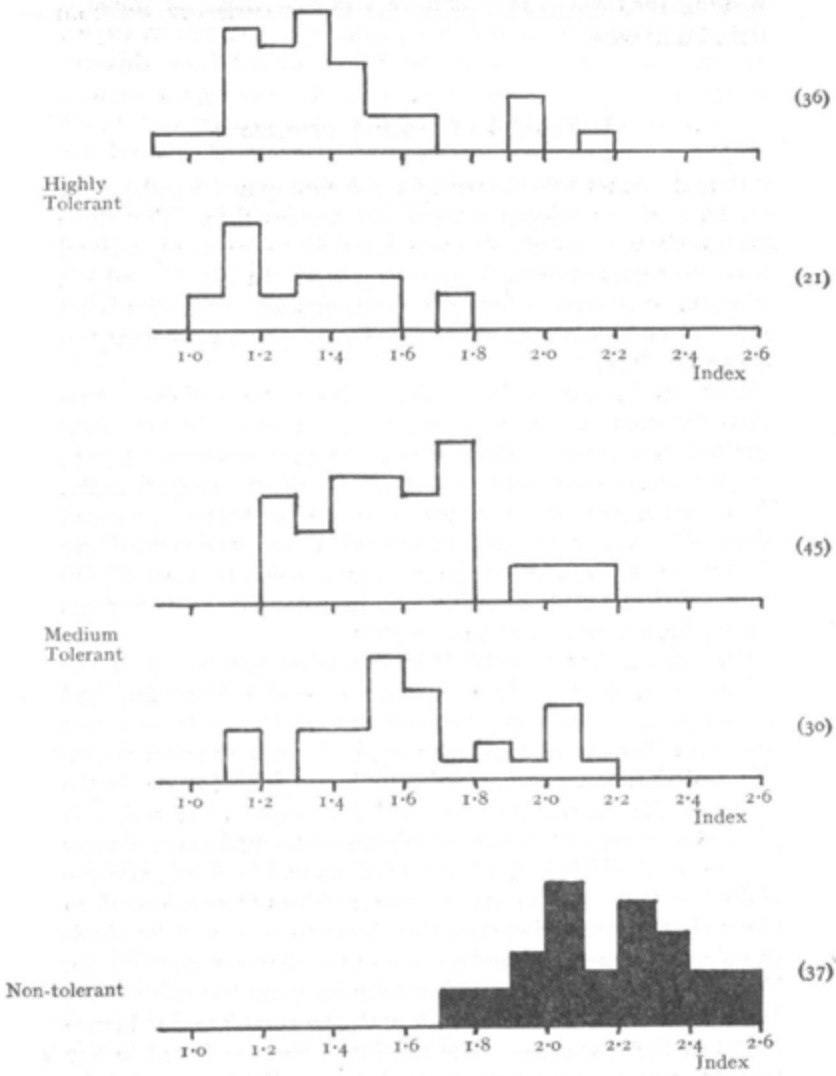

FIG. 2.—DISTRIBUTION OF LEAD TOLERANCE INDEX WITHIN FIVE POPULATIONS FROM LEADHILLS.
(Number of plants shown in brackets.)

further evidence about the genetic mechanism involved we must turn to the results of testing the progenies raised from artificial crosses.

5. Results of testing progenies

Parent plants were chosen in the first place on the joint evidence of the tolerance tests and species lists. The three grades already postulated were assumed to be valid, and all possible crosses were made between them. In Fig. 3A and B a selection of typical histograms of parents and progenies is set out. Wild populations which were certainly non-tolerant are shown in black.

Few results are so far available from the diploids. One cross between a highly tolerant and a non-tolerant plant resulted in a progeny whose mean tolerance was high (A : 749 × 137), and whose constituent plants did not differ significantly. This dominance of tolerance over non-tolerance appeared repeatedly, and when coupled as in this case with lack of any evidence of segregation in the progeny it could be most simply interpreted in terms of a single major gene—the two parents having been presumably homozygous.

One of the few successful diploid selfs gave proof of the existence of true medium tolerance in the Pennines, and suggested that the first interpretation of Fig. 1 (Means and Standard Deviations) was too simple. Plant 847 when originally tested had a medium tolerant index (1.60), close to the mean of the population from which it came. On selfing it yielded a progeny whose mean tolerance was again very similar —1.53—and which had a big spread, as will be seen from the histogram (B). Unfortunately, the numbers were too small to allow the differences between the plants to be tested for significance, but there appeared to be clear evidence of segregation. This suggestion that the medium tolerant plant 847 might have been heterozygous did not fit in with the complete dominance found in the first cross. The medium tolerance found in 847 and its progeny must have been due to a different gene from that conferring high tolerance on the progeny of 749 × 137. If the complete dominance found there was genuine, the attractive idea that the heterozygote might be medium tolerant must be ruled out.

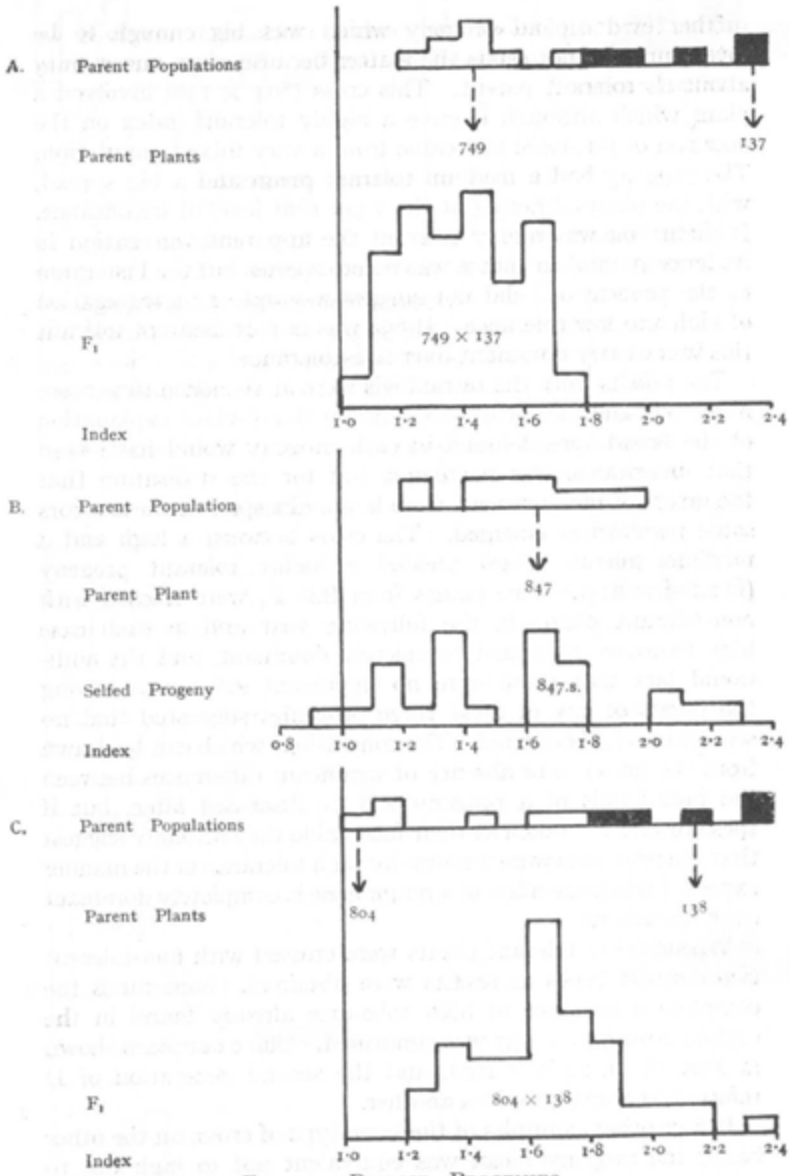

FIG. 3A.—DIPLOID PROGENIES.
(Non-tolerant parental populations are shown in black.)

A. High Tolerance dominant over Non-tolerance.
B. Medium Tolerance segregating on selfing.
C. Highly Tolerant parent with Medium Tolerant progeny ? (See text.)

The third diploid progeny which was big enough to be interesting did not settle the matter because of an uncertainty about its tolerant parent. This cross (804 × 138) involved a plant which although it gave a highly tolerant index on the occasion of its single test came from a very mixed population. The progeny had a medium tolerant mean and a big spread, with the plants differing at the 1 per cent level of significance. If plant 804 was highly tolerant the apparent segregation in its progeny implied that it was heterozygous, but the histogram of the progeny (C) did not suggest a simple 1 : 1 segregation of high and low tolerance. If 804 was in fact medium tolerant this was clearly dominant over non-tolerance.

The results from the tetraploids were at the same time more numerous and more complex. Again the obvious explanation of the broad spread found in each progeny would have been that inheritance was polygenic, but for the realisation that the errors of measurement were large. In spite of these errors some regularities emerged. The cross between a high and a medium tolerant plant yielded a highly tolerant progeny (D : 483 × 314). Two plants from this F_1 were crossed with non-tolerant plants in the following year and in each case high tolerance remained completely dominant, and the additional fact that there were no significant differences among the plants of any of these three progenies suggested that no segregation had occurred. The conclusions which can be drawn from the presence or absence of significant differences between the individuals of a progeny will be discussed later, but if these results are taken at their face value they strongly suggest that the progenies were uniform for high tolerance in the manner expected when one allele of a major gene is completely dominant over the other.

When highly tolerant plants were crossed with non-tolerant two distinct types of results were obtained. Sometimes the complete dominance of high tolerance already found in the diploid cross 749 × 137 was confirmed. One example is shown in Fig. 3b (E : 485 × 197), and the second generation of D referred to above provides another.

In two other examples of the same type of cross, on the other hand, the progeny mean was equivalent not to high but to medium tolerance, and at the same time there appeared to be some segregation. When an average plant from one of these progenies was backcrossed to a non-tolerant plant (F : 490

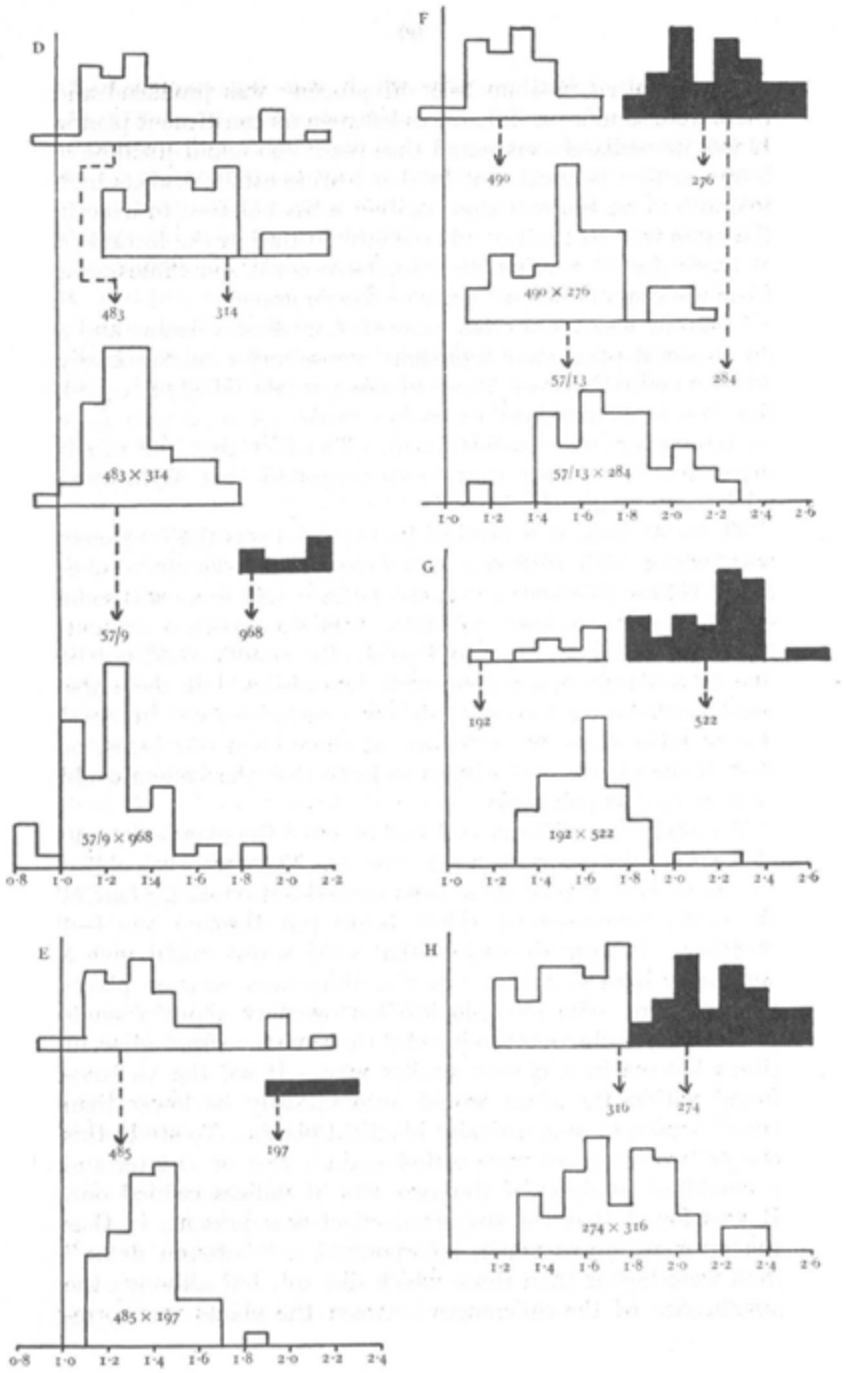

FIG. 3B.—TETRAPLOID PROGENIES. (D–H.)
Parent and progeny histograms arranged as in Fig. 3A.

× 276) another medium tolerant progeny was produced and there were significant differences between its constituent plants. It was immediately suspected that plant 490 might itself have been medium tolerant, but further testing established its high tolerance, and the fact that another cross behaved in exactly the same way (G : 192 × 522) confirmed that in the tetraploid at any rate the medium tolerant form could sometimes arise from the cross between the two extreme grades.

The only successful cross between a medium tolerant and a non-tolerant plant gave a progeny whose mean index actually fell somewhat between those of the parents (H : 274 × 316), but the high standard deviation made its departure from medium tolerance non-significant. The fact that the plants differed at the 1 per cent level suggested that segregation might have occurred.

The most notable feature of these results was the frequency with which high tolerance was found to be dominant over both medium tolerance and non-tolerance, and there was some evidence that medium tolerance was dominant over non-tolerance. It was also clear that all the results could not be due to a simple major gene with two alleles, but there was insufficient evidence to establish how many alleles were involved and whether these were present at more than one locus, or even if the number of loci was so large that the system could be described as polygenic.

The significant differences found between the plants of many of the progenies deserve some comments. These were calculated on the basis of two or three tests carried out on each plant at the same time—several tillers being put through the test together. It seemed possible that such a test might give a spuriously high significance to the differences between plants if there were some physiological homogeniety about a single plant which made roots produced at the same time from adjacent tillers behave in a closely similar way. If so, the variance found within the plant would automatically be lower than that between even genetically identical plants. To study this one or two progenies were tested again a year or so later and a combined analysis of the two sets of indices carried out. It was found that the suspected effect was present, in that the error variances which incorporated a " between dates " item were higher than those which did not, but although the significance of the differences between the plants was corre-

spondingly reduced it remained high enough for confidence that those differences had a genetical basis. In other words, the combined figures still suggested that segregation for lead tolerance was taking place.

The clearest piece of evidence that major genes might be involved was provided by those highly tolerant progenies within which no differences could be shown. If this meant that the differences observed were due purely to experimental errors in the test (and it is realised that such negative statements can never be properly substantiated), it implied that those progenies were genetically homogeneous. This would be expected with a major gene, whose dominant allele could exist in the homozygous form in a few plants even when a small population was surrounded by homozygous recessives, but it would not be expected with a polygene system. The alternative explanation that the apparent uniformity of the highly tolerant progenies was due not to any genetic mechanism in the plants but to some defect in the Index of Tolerance cannot be ignored. If a much lower concentration of lead had been used, so that any plant which was reasonably tolerant could grow with little interference, this is exactly the effect which would have been expected. In fact the concentration chosen normally halved the growth rate of the tolerant plants, and only a very few individual tillers gave index figures as low as the 1.0 which would indicate uninhibited growth. The possibility of extending the range of the index by using a higher and a lower concentration is still being considered, but it is not felt that so far the original index has shown any real evidence of inadequacy.

6. Conclusion

These somewhat inconclusive results are being reported here as an example of the difficulties of genetical analysis when the character of interest is difficult to measure. One definition of lead tolerance has been proposed and some of its properties investigated, but none of the small improvements in technique which have accumulated during the four years it has been in use have reduced the errors sufficiently to make it possible to establish the tolerance of an individual plant with accuracy. Some of the plants which have been used in a number of tests

are known reasonably well, so that highly tolerant, medium tolerant, and non-tolerant plants can be produced if required for further experiments, but for the bulk of both the original collections and the derived progenies only population means are known with any certainty. This makes it quite impossible to attempt to calculate mendelian ratios, or even to be sure that the ratios expected from a major gene system are there to be found. It may yet be possible to refine the existing technique enough to reduce the errors to a manageable size, but some quite different index, such as one derived from the effect of lead on the activity of a chosen enzyme in the root cells, may in the end be found more useful.

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THE DEME TERMINOLOGY

J. S. L. GILMOUR
University Botanic Garden, Cambridge

When the Director suggested that I might write a brief article for the Report on the deme terminology that he and I first put forward in 1939 (Gilmour and Gregor, 1939), and which Professor J. Heslop-Harrison and I (1954) have since developed more fully, I was grateful for the opportunity of bringing it to the notice of those readers of the Report who may not have seen the original papers.

The initial stimulus to produce such a terminology arose from the conviction that the multitude of categories which, by the 1930's, had been proposed for the units of Experimental Taxonomy (alternatively known as Biosystematy and Genecology) contained the seeds of much potential confusion, both as regards the *purpose* of the categories and the *overlap and inter-relationships* of the actual terms used.

The need for such categories stems, of course, from the great development of genetics and cytology which began about the beginning of the century, triggered off by the re-discovery of Mendel's work in 1900. This development gave biologists, for the first time, some knowledge of how micro-evolutionary change actually takes place, and necessitated the creation of a terminology to describe the units of such change. To take one example, Turesson, in Sweden, discovered, by his transplant experiments, that many of the slightly differing populations within a species were genetically distinct and had almost certainly arisen by selection in response to differing ecological conditions. He proposed the term ' ecotype ' for populations of this kind, and much subsequent work by Gregor, Turrill, Marsden-Jones, and many others, has confirmed that this type of micro-evolutionary change is a widespread phenomenon. Again, the inter-fertility and inter-sterility of populations, in nature and in the experimental ground, is obviously an important factor in their evolution, and categories (such as Danser's Commiscuum, Comparium, and Convivium) were proposed to describe units based on this factor. Gradually, a multitude of categories accumulated ; a duplicated report on

them compiled a few years ago by Sylvester-Bradley for the Systematics Association ran to nearly twenty pages !

One important feature of these categories is that the relationship between them and the categories of ' orthodox ' taxonomy (genera, species, &c.) has very rarely been clearly defined by the authors who put them forward. Are they intended to be additional to, or substitutes for, the ' orthodox ' categories ? This doubt is accentuated when the word ' species ' is incorporated into an experimental taxonomic term, as it is, for example, in Turesson's ' ecospecies '.

It is the contention of those putting forward the deme terminology that this is a key question, and that no satisfactory answer can be found without giving some thought to the nature and purpose of classification in general, whether applied to living or non-living things. This approach has been developed elsewhere (Gilmour, 1951) and in this brief article I will make only the following points :—

(1) Every classification should be judged in relation to the *purpose* for which it is made. No classification should be regarded as an end in itself, but as a *tool* to serve a particular need or needs. To say that one classification is " better " than another, without considering *" better " for what purpose,* is to make a virtually meaningless statement.

(2) It is necessary to make different classifications for different purposes. Thus, one makes a different classification of mountains if one is interested in their heights from that made if one is interested in their geology.

(3) In the case of some objects, it is possible to make *one* classification that serves a large number of purposes, in addition to making many other classifications, each serving a particular purpose. The possibility of doing this depends on there being *one* factor influencing the attributes of the objects that is more powerful than any other, thus enabling one to make a classification whose classes contain objects having many attributes in common. For example, in classifying mass-produced motor-cars, the powerful influence of mass-production enables one to classify the individual cars into a number of separate ' models ', each comprising cars with practically all their attributes in common. Such classifications (of which mass-produced car ' models ' form an extreme example) are termed, in the general theory of classification, *natural* classifications, and are useful for

a wide range of purposes, as opposed to *artificial* classifications (*e.g.*, all cars delivered to a certain town), which class together individuals with very *few* attributes in common, and which are useful virtually only for the purpose for which they were made. Thus, to know that a car belongs to a particular model enables one to predict its price, its speed, its comfort, and many other attributes, whereas to know only that it was a car delivered to Cambridge, is useful almost exclusively for purposes of Cambridge trade statistics. Intermediate classifications would be those into *types* of vehicle (*e.g.*, lorries, sports cars, &c.) or those based on *age* (*e.g.*, vintage cars, &c.)—intermediate in the number of common attributes, and in usefulness.

If we try to apply these general principles to the classification of living things at or about the species level, I suggest that we find a situation in which, owing to the powerful influence of heredity, it is possible to make *one* classification that is a natural one in the above sense and is, therefore, useful for a wide range of purposes, namely the classification into the orthodox taxonomic categories of species, subspecies, varieties, &c. Such a classification has not necessarily any micro-evolutionary purpose and, indeed, was made long before the fact of evolution was accepted. In order to continue to be useful for a wide range of purposes, it should remain as stable as possible and should be exempt from constant alteration, for either taxonomic or nomenclatural reasons. It should form, as it were, a broad map of the, mainly morphological, variability of living things, a map that is useful alike to the general and applied biologist, to the ecologist, the anatomist, the horticulturist, and the farmer. In addition, however, for special purposes, other classifications can be made, quite distinct from the ' orthodox ' natural classification, and one such classification can be a classification specifically constructed to study the mechanism of micro-evolutionary change. A difficulty arises here, that has, I suggest, led to much of the confusion surrounding the terminology of experimental taxonomy. In many organisms, especially among animals, the orthodox categories of species, subspecies, &c., based primarily on the correlation of morphological characters, coincide pretty closely with a grouping according to the factors, such as inter-sterility, of importance in micro-evolutionary change, and this coincidence has obscured the desirability of keeping the two classifications

distinct. This situation, however, by no means always occurs, and, especially in plants, one often finds, for example, that inter-sterility barriers cut right across a grouping according to morphological similarity.

It is, I suggest, essential, if confusion is to be avoided, for the two classifications, one for a wide range of purposes, and one for the particular purpose of studying the units of micro-evolutionary change, to be kept distinct, both in aim and in terminology. This is the basic point that led to the formulation of the deme terminology, a terminology designed, at the same time, to underline the separation of broad-purpose and micro-evolutionary classifications, and to provide a uniform and workable terminology for the latter.

The full range of terms suggested is contained in the paper published in *Genetica* in 1954; here I will give only the principle on which it is based and a selection of the terms included. The essence of the terminology is the construction of a series of category-terms by the addition of one or more virtually self-explanatory prefixes to the 'neutral' suffix "-deme". The suffix -deme is defined as "a term, always used in this terminology with a prefix, denoting any group of individuals of a specified taxon". (The use of the phrase "specified taxon" indicates the way in which the 'orthodox' categories can be used as a 'framework' into which other classifications, for particular purposes, can be fitted.) An important point to note is that the suffix -deme is *neutral*; that is to say, it carries no implication that the individuals exhibit any relationship other than that they belong to a specified taxon. In particular, it carries no implication that they form a *population*, in either a topographical or an interbreeding sense; these, and other implications, are indicated in the terminology by *prefixes*.

The ten basic terms suggested, with their definitions, are as follows:—

topodeme: a deme occurring in a specified geographical area.
ecodeme: a deme occurring in a specified kind of habitat.
phenodeme: a deme differing from others phenotypically.
genodeme: a deme differing from others genotypically.
plastodeme: a deme differing from others phenotypically but not genotypically.
gamodeme: a deme composed of individuals which are so situated spatially and temporally that, within the limits of the breeding system, all can interbreed.

autodeme : a deme composed of predominantly autogamous individuals.

endodeme : a gamodeme composed of predominantly endogamous (*i.e.*, closely inbreeding) dioecious plants (or bisexual animals).

agamodeme : a deme composed of predominantly apomictic plants (or asexual animals).

clinodeme : one of a series of demes which collectively show a specified variational trend (*i.e.*, which collectively form a cline).

Subsequently, the term **cytodeme** has come into use for a deme showing chromosome differences from other demes.

Reference can be made to the *Genetica* paper for a full explanation of these terms, together with examples, but a word may be said here on the term ' gamodeme ', which is essentially synonymous with the phrase " breeding population " as employed in micro-evolutionary and genetical literature, and hence represents a very basic concept. The term has been taken up to some extent by biologists, but in some cases (*e.g.*, in Huxley (1942) and Carter (1951)) the bare suffix ' deme- ' has been used in place of it, no doubt partly because it is shorter. This usage, it should be emphasised, cuts across the whole idea underlying the deme terminology, as it is essential to keep the suffix -deme completely ' neutral ', otherwise the connotation of " breeding population " becomes injected into *all* the compound terms, thus destroying the intended use of many of them. For example, the term ' ecodeme ' is designed to cover *all* individuals of the taxon occurring in a particular type of habitat, right throughout the range of the taxon, and carries no implication that such individuals form one interbreeding population.

In addition to the basic, ' first order ', derivatives, ' second order ' derivatives can be constructed, and about two dozen are suggested in the *Genetica* paper. Examples are :—

genoecodeme : an ecodeme differing from others genotypically.

hologamodeme : a deme composed of all of those individuals which, within the limits of the breeding system, are believed to be able to interbreed with a high level of freedom under a specified set of conditions (*cf.* TURESSON's " ecospecies ").

coenogamodeme : all hologamodemes considered to be capable of exchanging genes through their members to some extent, but not with freedom, under

a specified set of conditions (*cf.* TURESSON's " coenospecies ").

syngamodeme : all coenogamodemes connected by the ability of some of their members to form viable but sterile hybrids under a specified set of conditions (*cf.* DANSER's " comparium ").

' Genoecodeme ' corresponds broadly with Turesson's ' ecotype ', and the last three are designed to cover the most important of the situations commonly encountered in the investigation of the fertility relationships of populations.

One point was not perhaps stressed as much as it should have been in the *Genetica* paper, and that is the question of the naming of *individual examples* of the various categories. Some biologists have asked if it was intended that these should be named according to some definite and uniform system—perhaps with Latin names parallel to those of the taxonomic categories. This was certainly not the intention. In some groups, of course, deme categories will coincide with taxonomic categories ; for example, genoecodemes with subspecies, and the genoecodemes will then, in effect, have a Latin name *as a subspecies*. It was the intention, however, that the deme categories, *as such*, should be referred to by a loose, *ad hoc*, method of nomenclature, varying with, and adapted to, the particular type of investigation being carried out. For example, if a series of gamodemes in a particular area is being studied, each gamodeme could be referred to by a number or letter, or by a topographical adjective if that were more appropriate.

The above very brief outline will, I hope, give a broad idea of the aim and scope of the deme terminology. Its adoption, in whole or in part, will depend, of course, on the acceptance of the thesis that a ' natural ' classification is distinct in aim, and should be distinct in terminology, from a micro-evolutionary classification. This point is a fundamental one, and there are signs that it is receiving consideration from biologists ; Cain (1959, p. 316), for example, has recently discussed it in relation to the difference between the taxonomic practice of zoologists and botanists. If the deme terminology can claim to have played a part in stimulating this interest, its proposers will feel that it has served a useful purpose.

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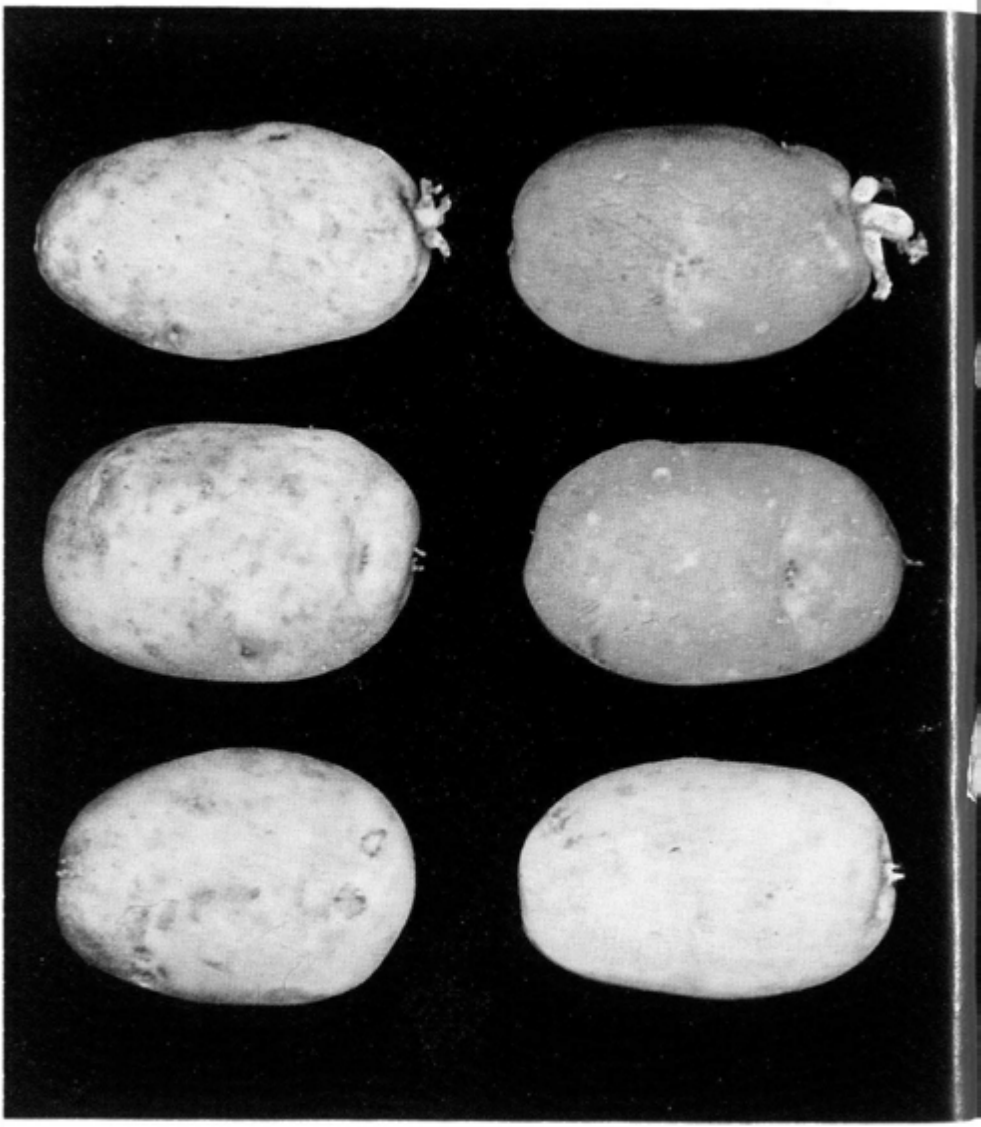
POTATOES BRED AT THE PLANT BREEDING STATION

WITH TWO RED VARIANTS



Top : Craigs Royal; Red Craigs Royal

Middle : Pentland Beauty; Red Pentland Beauty

Bottom : Pentland Crown; Pentland Dell