THE ECOTYPIC DIFFERENTIATION OF POPULATIONS: ALTITUDINAL DISTRIBUTION OF VARIATION IN *FESTUCA*.

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I. INTRODUCTION.

Towards the end of last century and, indeed, before ecology cytology and genetics had attained any real status, interesting experiments were carried out from the point of view of the influence of the environment upon plants. The most important were those of Kerner who published an extensive account (1891) of his investigations on the reactions of both annual and perennial plants at different altitudinal stations. He came to the conclusion that a distinction must be drawn between modifications and hereditary differences and emphasised that the former cannot be transmitted by the parents to their offspring. In France Bonnier set up a series of experiments at lowland and alpine stations and must be acknowledged as the first investigator to introduce the use of clonal transplants. He published a final account (1920) of the experiments which he had conducted over a very long period but his results must be regarded as dubious if only because his "stations" were untended save at irregular intervals and, being in the wild, were subject to the natural contamination of such conditions. In America, Hall (1932) was the first to recognise the importance of an experimental taxonomic approach and his work has been amplified and extended on a large scale by

Clausen, Keck and Hiesey (1940) who have also made use of many of the ideas propounded by Turesson (1922) who originated the method of growing plants from different climates under the uniform conditions of a garden. The name of Turesson will remain closely linked with this method of investigation and with the units of experimental taxonomy which he first introduced into scientific literature. In Britain the most important work in this field has been done by Gregor on Plantago maritima. Gregor, Davey and Lang (1936) and Davey and Lang (1939) have set a very high standard of precision for this type of work and Gregor (1938, 1939, 1946) has come to conclusions of vital importance to the understanding of plant communities. The clonal transplant experiments of Marsden-Jones and Turrill (1930, 1933, 1935, 1937) at Potterne have been of great value in a rather different way for they have enabled the ranges of edaphic tolerance of individual genetic constitutions to be assessed.

This experimental study of natural populations has led to a fuller appreciation of the relationships between local communities and to the recognition of the factors which contribute to their formation and composition. The area of investigation has in a sense been narrowed but the taxonomic implications of the findings are of even greater fundamental importance and are no longer the interest of the systematist alone. The results of such investigations cannot be described in the language of orthodox taxonomy and the ecotype is but one of the units which have been defined in an attempt to create a flexible system of population classification which will be, in a very real sense, complementary to that of the systematist.

The true differentiation of populations has its origin in hereditary variation. Modificatory variation is the morphological response of the individual to the prevailing conditions of the environment and is of little interest from the evolutionary point of view since there is no question of such adaptations being inherited by succeeding generations. It has to be remembered that modifications are almost invariably present in plants growing in the wild and though they may only serve to exaggerate the hereditary differences, they may also distort them. On the other hand, hereditary variation is the direct expression of the peculiar constitution of the genetic constitution of the individual and is, therefore, of primary evolutionary importance.

A careful consideration of differentiation reveals the fact that the same factors are operative whether it is the species, the population or even individual organisms which are under review. It also becomes clear that though it may be more simple to grasp the importance of the factors at the higher taxonomic levels, it is at the lower levels where some apparent confusion exists, that a real understanding of their working may be gained. The fundamental source of variation within the genetic constitution of the organism is constantly providing new material through mutations and through chromosome changes. The mutation rate, then, of a species must provide a clue to the possible amount of variation within it but gives no indication of the actual extent since other factors also play an important part. Natural selection is perhaps the factor which most readily comes to mind.

It has been thought of as a sieve which eliminates non-viable mutations but Sinskaja (1942) criticises this idea since, she says, "each individual heritable change ..... becomes incorporated in the entire constitution of a plant organism." This cannot be altogether correct since it is obvious that lethal mutations preclude survival in any form and that those which are harmful render the plant more liable to elimination although it is true that those constitutions which are not too closely adapted to a given environment are most capable of successful survival under changed conditions.

The extent and nature of the variation is, however, dependent also upon the size of the population and in this connection it is worthy of notice that the effective size is limited to the number of mature individuals. Sewall Wright in particular has pointed out the importance of this question of the size of populations and emphasised (1940) that with small numbers "selection pressure becomes ineffective while mutation-pressure is not affected." Variability is in time reduced in such populations with few breeding individuals and the chance fixation of even harmful mutations is a possibility. In general, the tendency is towards a random drifting apart from neighbouring communities and as the reservoir of evolutionary change is diminished, inbreeding may lead to the extinction of a small isolated population especially under stress of changing conditions.

An illustration of this process at work in nature may be taken from an account by Sinskaja (1931) of the wild radish of the sea-coasts of Japan. She notes that the habit conditions favourable to the growth of this plant are being destroyed through the agency of man; the number of habitats are decreasing and the area is being severely restricted. Under these circumstances she says "an impoverishment as to forms has indubitably taken place" and thinks it possible that the species will in time cease to exist.

The environment is never uniform throughout the area of distribution of a species so it follows that even where there is a high degree of continuity, local populations may become differentiated in some degree in response to the selective action of habitat conditions. Where gene flow is uninterrupted in spite of partial discontinuities, such large populations provide a most important reservoir of potential evolutionary change but it is also understandable that with the occurrence of definite ecological conditions, changes may take place within the genetic constitution of the habitat community which form a barrier to the free exchange of genes with neighbouring populations. Where inter-breeding between such communities is not impossible, hybridisation frequently occurs only in a comparatively narrow zone between discrete populations. That this hybrid weakness is probably due to a loss of stability or harmony within the gene-complex rather than to any selective action accountable to the environment may be deduced from data collected by Sumner (1932) on Peromyscus subspecies in Florida. P. p. albifrons is a pale type which inhabits the white sand beaches whilst the inland subspecies P. p. polionotus is dark in colour and found on an area of dark soil. There is a continuous gradient of pelage-colour change which is gradual until about forty miles inland where it becomes very steep for a short distance and then resumes its gradualness. It appears that this narrow zone is the boundary between the two subspecies but the interesting point is that it does not coincide with the boundary between the two soil types. The suggested explanation is that pressure of an increasing population has caused migration outwards from the P. p. polionotus area and thus extended the boundary of the subspecies beyond the soil type change. It seems, therefore, that though hereditarily adapted for a different environment and, therefore, sustaining some loss of protection against predators, the migrants are nevertheless more capable of survival than are the hybrids which must suffer some loss of gene-complex stability. The division between populations is not always so abruptly marked, however, and in cases where the intra-population balance is not so finely adjusted, it is much more common to find a gradual intergradation between the extremes.

The fact that the environment has an effect upon the morphological characteristics of a species has long been appreciated and a change from tall to dwarf plants is expected along a range from fertile, sheltered areas to exposed areas of poor soil. Such changes may be due entirely to modifications but if they can be proved to be hereditary, interest is concentrated not on the morphological attributes involved so much as on the direction of change, its continuity and the probable initiating factor.

It is, indeed, the gradual intergradation which is of particular interest and which is examined in the present work. There is a considerable volume of literature which shows that the variation which may be observed under natural conditions is not haphazard and unrelated but the result of the interaction of a varying number of factors. The analysis of this variation together with a consideration of the factors involved make it possible to assess the importance of the contributions made by different sources and causes of change and also the degree and direction of the differentiation. It may be found that trends in character variation are not always parallel and that in some cases the gradients cross each other at an angle but it is important that they should be recognised for only in this way will it be possible ultimately to present a picture of the internal hereditary structure of the population as a whole.

The fescue species which are the subject of the present investigation are not in themselves of economic importance but it was considered that an appreciation of the trends which occur within such widely distributed species could provide a useful basis for the understanding of the much more complicated position which exists within species of crop plants.

It has been calculated that the temperature falls approximately 3°F for every 1000 feet of altitude "so that all mountains reproduce, according to their height, a temperature range corresponding to that between certain latitudes at sea level" Good (1947). The mountains of Great Britain are not sufficiently high to reproduce a very wide latitudinal range but within the limits of this country, an altitudinal gradient provides a fairly varied series of environmental conditions. The study of character differentiation requires the collection of representative samples from as many habitats as possible and the choice of an altitudinal gradient enabled adequate samples to be obtained in a relatively short time.

II. THE SAMPLING AREA.

Ben Cruachan in Argyll (lat. 56° 25', long. 5° 8') was chosen as the source of the material for the present investigation, the chief reason being that the vegetation which covers the greater part of the mountain, is essentially of the grassland type and is comparatively uniform. Again a preliminary survey of the area in 1944 showed that fescues occur in almost unbroken sequence from sea level at Loch Etive to the summit (3689 feet), a reasonably extensive range to obtain in the mountains of Great Britain.

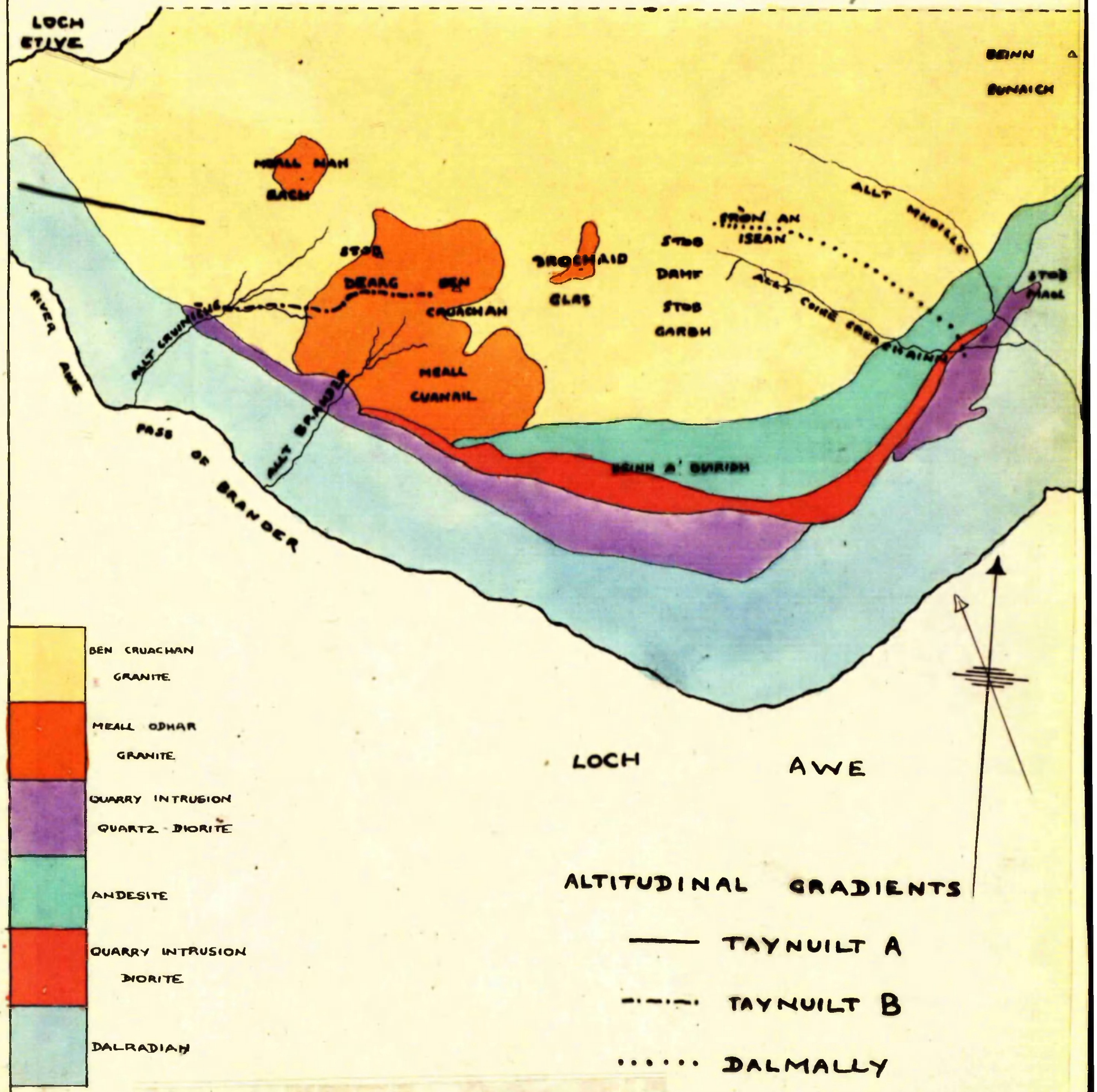
Ben Cruachan is an uncultivated granite mass in which the most widespread type of rock is a grey hornblende-biotite-granite of fairly coarse grain though a slightly more acid pink granite does occur at the highest altitudes (Figure 1). A belt of andesitic rock bordered to the south by a band of diorite, cuts the granite in the south and east but the chemical analyses show little significant difference (Anderson, 1937). A zone occurs on the perifery of the granite where the older sedimentary rocks show the effect of contact metamorphism but it is of small importance in the present investigation. Porphyritic dykes are fairly frequent in the granite mass and might be expected to cause differences in the pH of the soil. Samples of soil, however, were tested with the BDH Soil Indicator and all had a pH ranging Between approximately 4 and 5.5 with the exception of one sample from the Cruachan Quarries which occur on the Quartz-diorite and rather surprisingly, one from the summit. The comparative uniformity of the acid

FIGURE I

FIGURE I

PLAN OF CRUACHAN MASSIF

GIVING GEOLOGICAL DATA



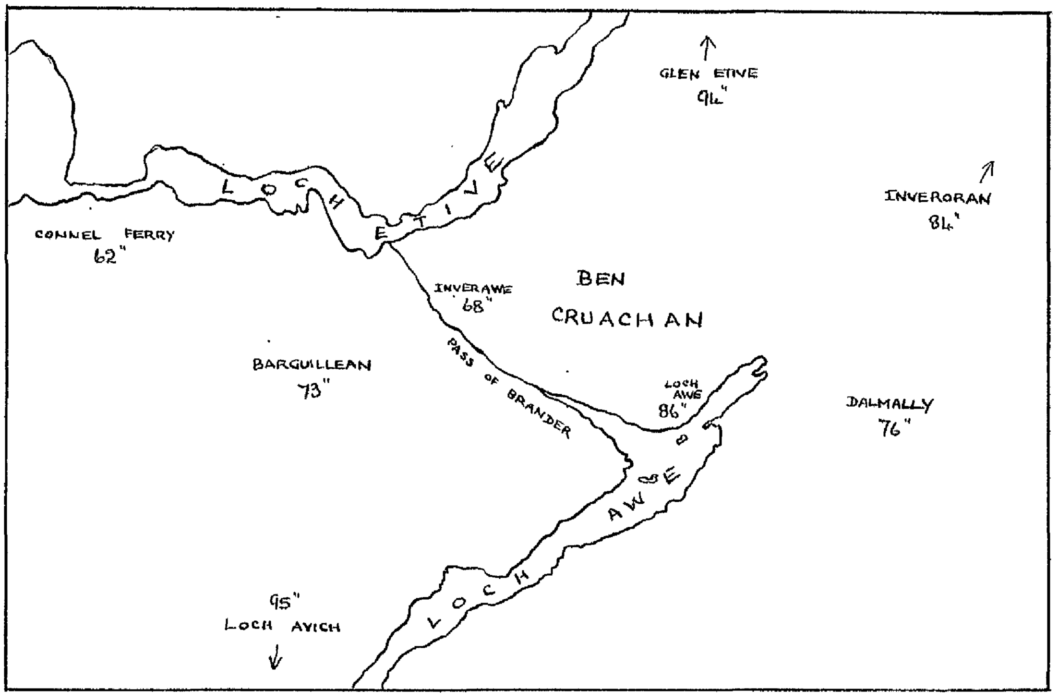
reaction is probably due to the fact that the underlying rock is almost everywhere covered by a layer of peat of varying depth.

The area at sea level on the Taynuilt side of the river Awe is geologically quite distinct from Ben Cruachan as it is a raised beach. The coarse pebbly sand of the shore gave a neutral reaction with the BDH Soil Indicator and so did the soil in crevices of a sheltered mortar-built wall but all other soil samples showed an acid reaction.

No records of the actual rainfall on Ben Cruachan are available but annual averages were obtained by courtesy of the Meteorological Office for various weather stations in the immediate vicinity.

FIGURE II.

Weather Stations in relation to Ben Cruachan.
Annual Rainfall Averages



It has been suggested that, taking into consideration the position and aspect of the mountain and the records for similar mountain masses, it may be calculated that the average annual rainfall will increase from 80 inches at the Pass of Brander to at least 110 inches at the summit.

An examination of the average monthly rainfall for the Loch Awe, Cruachan, weather station shows that June and July are the driest months of the year. Nevertheless, mist frequently lies on the Ben at least as low as 2000 feet for considerable daily periods throughout the year including these months and though it may not add much to the rainfall, it must have the effect of reducing transpiration to a minimum and is therefore a factor of considerable importance.

No observations are available for wind velocity but some general statements may be made which at least indicate the trend to be expected. Chapman (1919) shows that there is a fairly rapid increase in velocity up to 500 metres (1640 feet) in respect of light winds and up to 300 meters (984 feet) in respect of strong winds. Thereafter the increase is generally less rapid but also more erratic. These observations refer to the atmosphere but it may be assumed that some comparable but less rapid increase in velocity takes place along a rising land surface. Durward (1921) noted diurnal variation in wind velocity at different heights and the main point of interest is that with all winds there is a tendency towards a minimum velocity about mid-day. This fact may have some bearing upon the distribution of pollen since the anthers in the fescues have been observed to dehisce mainly between the hours of nine and eleven in the morning.

Two altitudinal gradients were chosen for the purposes of the investigation and the fescue populations along each were sampled at intervals of 200 feet (Figures I and II.). In addition several habitat stations were sampled at sea level on the Taynuilt side of the River Awe.

1. The Taynuilt gradient. Owing to the configuration of the mountain, this gradient is in two divisions; A, the lower with a westerly aspect extending from sea level at Loch Etive up the slopes of Meal nan Each to 1600 feet, B, the upper with a south-westerly aspect from 1800 feet above Allt Cruiniche to the summit (3689 feet).

2. The Dalmally gradient. This gradient extends from 400 feet at the Cruachan Quarries along the very even ridge between Coire Chreachainn and the valley of Allt Mhoille. It has a south-easterly aspect and is, therefore, sheltered from the prevailing wind which is west to south-west.

Marked similarities occur between the Taynuilt and Dalmally gradients and one of the most obvious is the presence of bracken at the lower altitudes. Extensive patches and belts are found on the Taynuilt A gradient to 500 feet but on the Dalmally gradient the bracken grows to over 1000 feet. The bracken is succeeded on Meal nan Each by a narrow belt of Molinia-Nardus grassland which very soon gives place to a more level, very wet area where rushes and marsh plants are frequent save on the many drier, grass-covered knolls. There is no equivalent area on the Dalmally gradient which is on the whole, well drained and where a Fescue-Nardus grass area becomes dominant as the bracken fails. At higher altitudes a similarity between the two gradients again

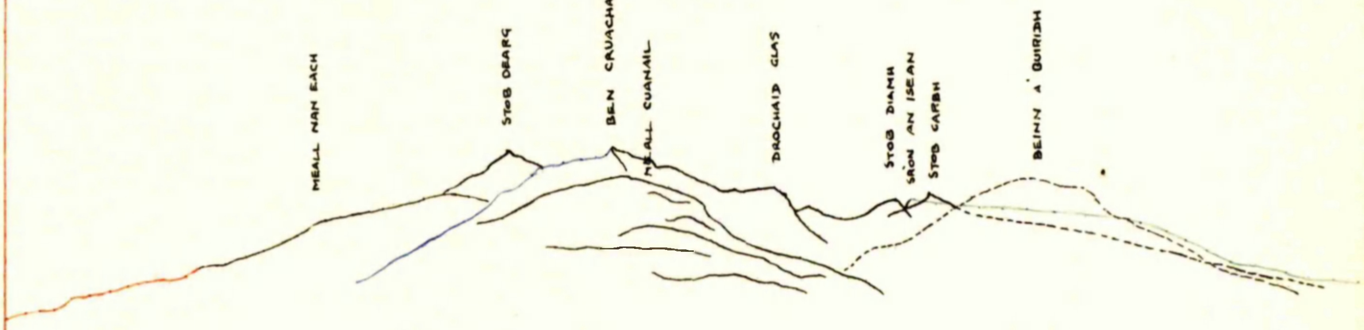
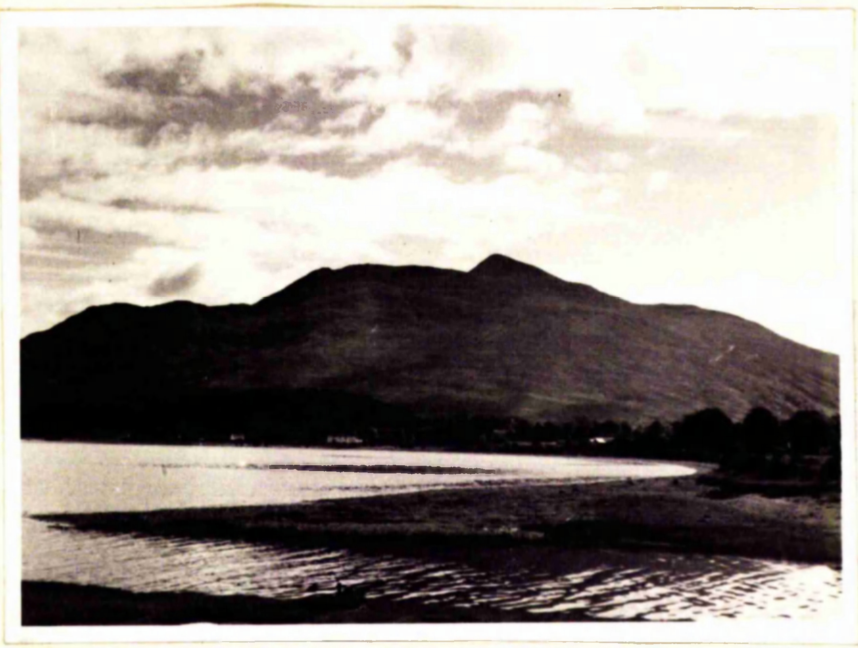
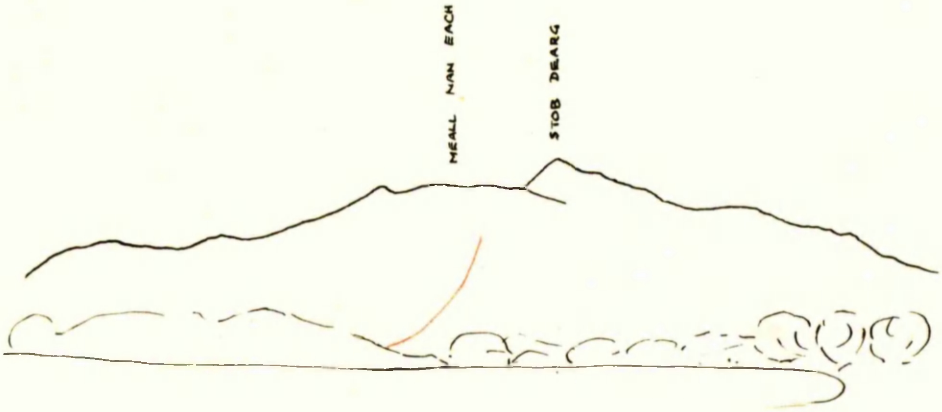


FIGURE III. ELEVATION OF THE CRUACHAN MASSIF SHOWING ALTITUDINAL GRADIENTS.

—— TAYNUILT A —— TAYNUILT B —— DALMALLY.





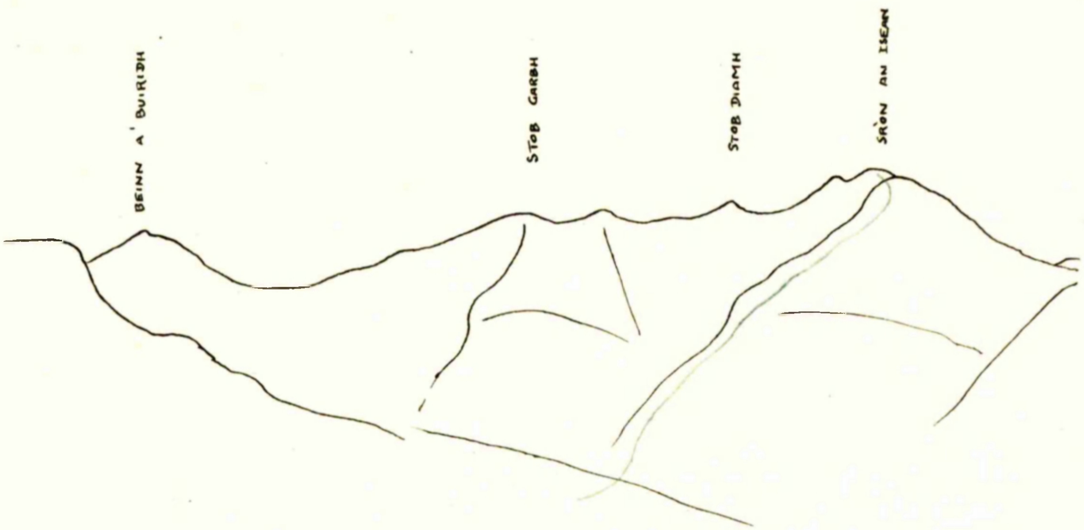
BEN CRUACHAN FROM AIRDS BAY. SKETCH OF TAYNUILT A.

appears with an extensive stony area of very poor, thin soil on the Dalmally gradient and a steeper, more rocky continuation of the Taynuilt gradient to 1600 feet. The B division of the latter gradient begins well above the bracken line, in Fescue-Nardus grassland and is replaced by an area of rock and sparse vegetation rather similar to the upper portion of the Taynuilt A gradient, as the altitude increases.

Sheep graze over the entire area of Ben Cruachan but cattle are also found at lower altitudes on the Dalmally gradient and on the Taynuilt sea-level raised beach.

Fescues at sea-level are abundant, relatively evenly distributed and tolerant of a fairly wide range of conditions, showing an ability to colonise habitats such as the open sea-shore and the top of a dry-stone wall where other plants seem unable to survive. At other stations at sea-level, fescues are growing under conditions of severe competition from grasses, mosses, bushes and even small trees and such habitats are sheltered. Sexual forms are predominant but viviparous forms were also found at two stations. This Taynuilt area is undoubtedly distinct in its environment and is separated from the Cruachan fescues not only by the River Awe but also by an area of cultivated ground and woodland.

The distribution of the fescues on the Ben Cruachan gradients is least continuous in the bracken areas but even the most dense growth of bracken may be broken by boulders and where these are large enough, it is possible to find a small colony of fescues or even a single plant growing in a crack or hollow. Fescues also occur wherever the bracken thins out sufficiently and it is



SKETCH OF CRUACHAN QUARRIES FROM DALMALLY SHOWING GRADIENT.

(BELOW) THE CRUACHAN MASSIF FROM ABOVE TAYNUILT.

obvious that though the fescue population is limited by the density and shading effect, the bracken does not provide an impassable barrier. Another obstacle to continuity of distribution is encountered in the very wet areas for though fescues may be found in such parts and have even been observed on small gravel islands in a stream bed, they do not appear to survive in those parts which are water-logged throughout the year except in unusually dry seasons and where, possibly, the limiting factor is the poor aeration of the soil.

In general, sexual and viviparous forms occur together at all altitudes up to about 2600 feet on the Taynuilt gradient whilst above this point, the sexual form disappears entirely and the viviparous form is undoubtedly the dominant in the mountain vegetation. Indeed, the sexual forms are most common at low altitudes, the viviparous alone at high altitudes and in the intermediate zone the sexual and viviparous are found in fairly equal proportions though the sexual forms are very sparse at some stations and tend to set seed late and consequently the amount of seed is small. The same broad picture is true of the Dalmally gradient though there the sexual fescues do not grow above 1500 feet and though the viviparous form is frequent thereafter, it does not become dominant until an altitude of 2100 feet is reached.

III. THE DELIMITATION OF CHROMOSOME RACES.

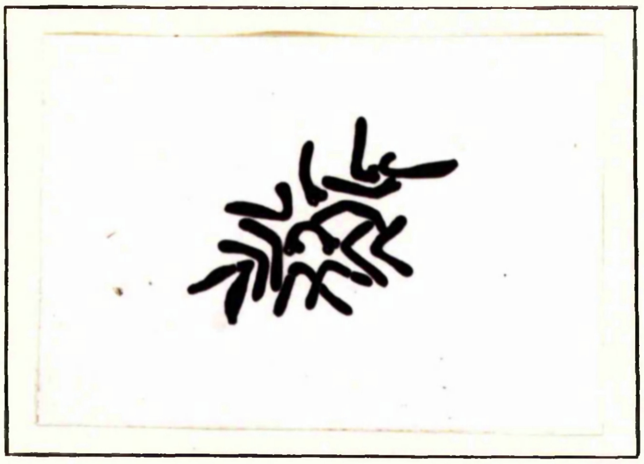
Various techniques were employed in an attempt to ascertain the optimum method of preparing root tips of the fescues for chromosome examination.

1. <u>The Squash Method.</u> Trials were carried out using acetocarmine squash technique (Belling 1926, McClintock 1929) and as modified by Thomas (1940). The staining, however, was not as satisfactory as that obtained using the Feulgen squash method (Darlington and La Cour 1942) and the acetocarmine method was consequently abandoned.

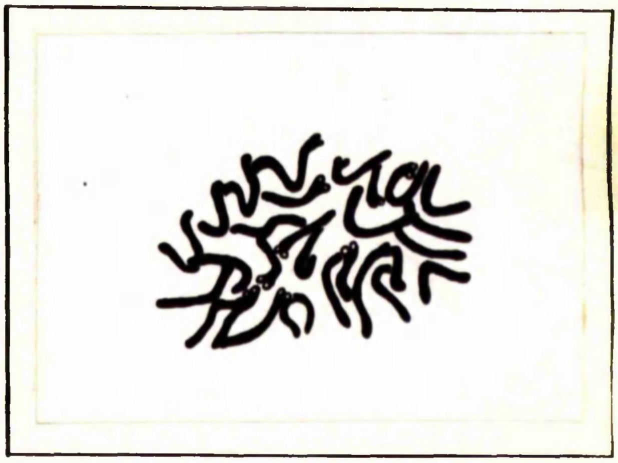
2. <u>The Paraffin Method.</u> The result of embedding and sectioning root tips was so much more satisfactory than the squash method that it was adopted for the entire investigation.

Lewitsky's fixing fluid (Lewitsky and Kuzmina 1927) gave a clearer result than an osmic acid fixative such as La Cour's 2BE (La Cour 1931) and modifications of Navashin (Müntzing 1933, Randolph 1935) were also used successfilly. It is possible that 2BE would have given the most satisfactory fixation had it been possible to find a reliable method of removing from the root tip some substance which became darkly stained by the osmic acid and invariably masked the cells of the periblem so effectively than an accurate count of the chromosomes was an impossibility. The usual method of using bleach to remove the osmic acid staining was effective for other parts of the root tip but was useless for these cells which are so important. The chromic acid in other fixatives also tended to darken this substance but the result was never so completely disturbing. The Feulgen stain was used until it was suggested that the hydrolysis in Hydrochlorin Acid could have the

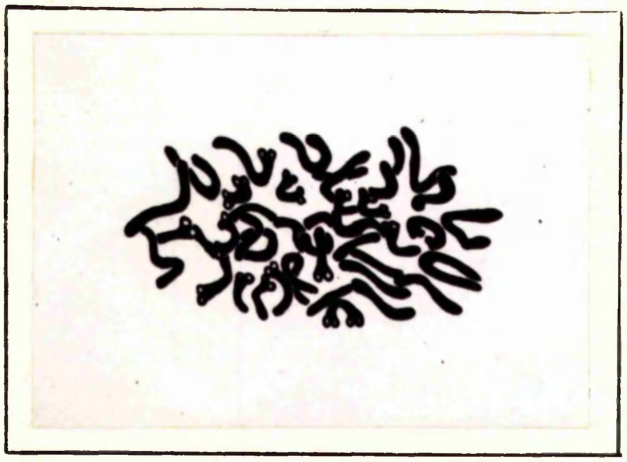
CHROMOSOME NUMBERS FOUND IN BEN CRUACHAN FESCUES.



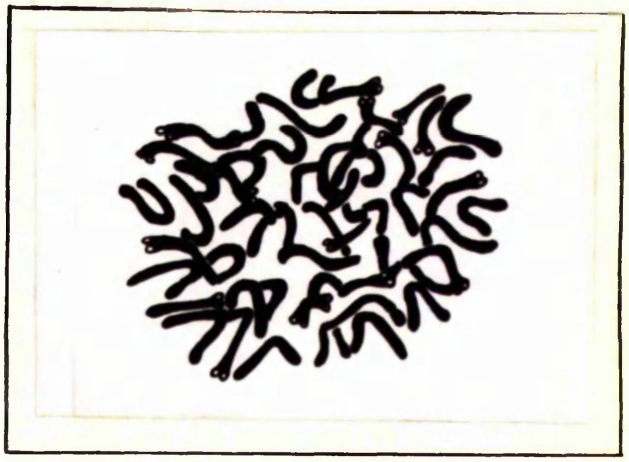
2n = 14



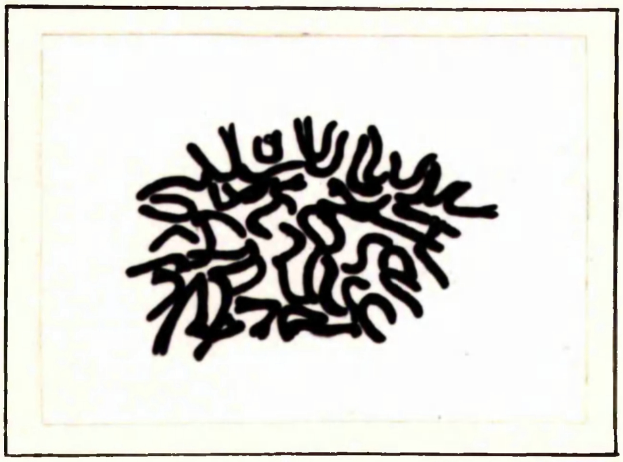
2n = 28



2n = 42



2n = 56



2n = 49

CAMERA LUCIDA DRAWINGS. MAGNIFICATION APPROX. × 1400.

effect of making the chromosomes brittle and might, therefore, cause fragmentation and thus render a count inaccurate. Crystal Violet was tried using the method suggested in Darlington and La Cour (1942) and found to be satisfactory but was afterwards replaced by Methyl Violet (Johansen 1943) which gave better definition.

Chromosome races.

Samples of the plants growing at each station on the two gradients were obtained and separated into two groups, (a) sexual and (b) viviparous. Root tips were taken from each plant and sections cut at 10μ. The prepared material was examined using a 1/12" oil immersion lens and X15 eyepiece with the following results (Table I).

TABLE I.

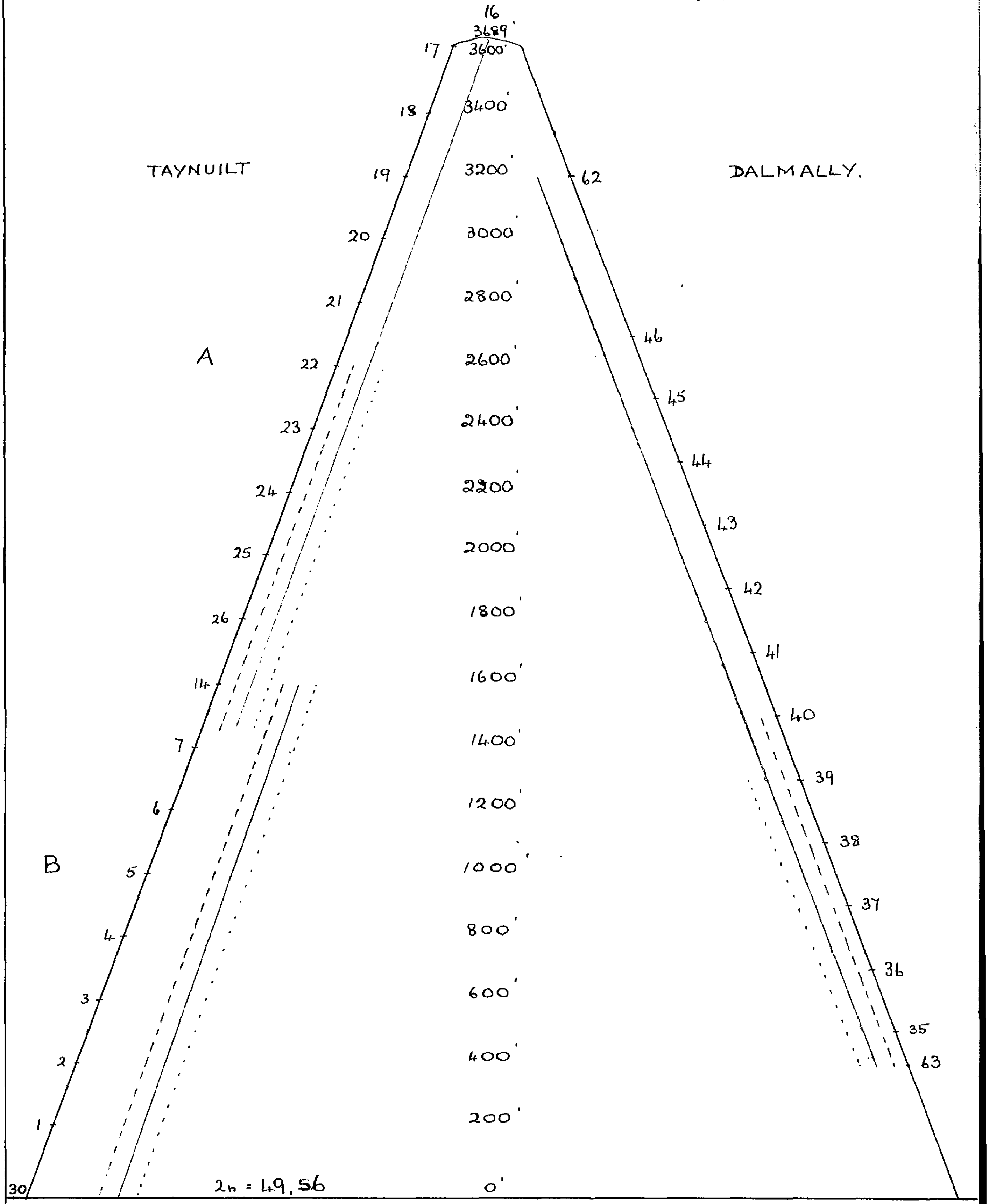
Chromosome counts in Ben Cruachan Fescues.

	(a) Sexual	(b) Viviparous
2n Chromosome Counts.	14 28 42 49 56	28

The large majority of the sexual population came under the two groups (2n = 14) and (2n = 42). The other counts recorded for sexual plants were all counted in samples from stations at sea level. The tetraploid 2n = 28 was represented by a single plant and the aneuploid (2n = 49) was almost equally rare occurring only twice but in different populations. On the other hand, the octoploids (2n = 56) represented a small population occupying a partially isolated habitat and including one of the

FIGURE IV

THE DISTRIBUTION OF THE CHROMOSOME RACES ON THE ALTITUDINAL GRADIENTS.



- - - - - - - - DIPLOID SEXUAL $2n = 14$

——————— TETRAPLOID VIVIPAROUS $2n = 28$

. . . . HEXAPLOID SEXUAL $2n = 42$

1 - 63, STATION NUMBERS.

occurrences of the aneuploid. As far as the viviparous forms are concerned all samples were alike in having the tetraploid number (2n = 28) (Figure IV.)

It is of interest to note that root tip sections from plants grown from seed collected at the same stations were also examined and the counts agreed largely with those obtained from the wild plants. Only one aneuploid count, however, was obtained. This would suggest that the aneuploid, which is very probably a hybrid between the common hexaploid (2n = 42) and the much rarer octoploid (2n = 56) is infrequent and, perhaps, does not produce much viable seed.

Jenkin (1934) has reported 2n = 28 for the sexual setaceous-leaved fescues and since the Ben Cruachan fescues of this type are all diploid (2n = 14) save the solitary tetraploid individual at sea level, it seemed necessary to endeavour to establish whether or not the prevalence of the diploid race on Ben Cruachan is in any way a chance phenomenon. Accordingly samples of setaceous-leaved individuals were collected from widely separated areas in Britain and examined for the presence of diploids and tetraploids. Again the sexual and viviparous forms were separated and the following results were obtained (Table II).

TABLE II.

Chromosome counts from a survey of setaceous-leaved fescues in Britain.

	Sexual		Viviparous
	$2n = 14$	$2n = 28$	$2n = 28$
Scottish Islands			
Shetland			x
Barra			x
Iona	x		x
Scotland West			
Inverness			x
Argyll Cruachan	x	x	x
Glen Coe			x
Scotland Central			
Perthshire Killin			x
Glen Clova	x		x
Scotland East			
Midlothian	x		
Berwickshire		x	
England North			
Yorkshire		x	
England South-west			
Cornwall		x	
Wales West			
Cardiganshire		x	
Montgomery		x	

An examination of Table II shows that the prevalence of diploids on Ben Cruachan is in no way exceptional but is in fact, typical of the northern region of Britain. Tetraploids do occur but they are always of the viviparous type with the exception of the sexual tetraploid at Taynuilt. The change in predominance from diploid to tetraploid in the southern part of Britain is striking and is worthy of a more intensive survey since it introduces an interesting biogeographical problem. From the present data it is impossible to say where exactly the change takes place.

IV. THE BREEDING STRUCTURE OF SEXUAL POPULATIONS.

1. Diploids. A number of trials were carried out under controlled greenhouse conditions to test the breeding relationships between populations. Plants from widely separated altitudinal stations were employed as well as from neighbouring stations and in all combinations, seed was set abundantly, the fertility percentages ranging from 11 per cent to 60 per cent. The seeds obtained from these matings were subsequently germinated on damp filter paper and yielded practically 100 per cent normal seedlings.

The self-compatibility of the plants used in these hybridisation tests was also investigated by reserving several panicles on each plant for self-pollination and the results obtained were in striking contrast to those for cross-fertilisation. The percentages of seed set ranged from 0 per cent to 3 per cent. These seeds were also germinated on damp filter paper and practically 100 per cent germinated but several produced albino seedlings.

It is obvious in the light of these results that in the wild, self-fertilisation is rare and that the seed which is produced is almost exclusively the result of cross-fertilisation. Moreover since members of populations from widely-separated altitudinal stations show no less ability to cross than individuals from adjacent populations, it may legitimately be assumed that there are no barriers of genetical origin, at any rate, which prevent a free flow of genes between populations throughout the area investigated.

A general picture of the distribution of the fescues in the area has already been given and to particularise concerning the diploid race, it may be added here that colonial development is not on the whole marked, although at the lower elevations, the distribution does not show strict continuity owing to the restrictions imposed by the phytosocial environment. Again, towards the upper limit of the occurrence of the sexual fescues, the density is reduced although the continuity is maintained.

2. Polyploids. Some trials were also carried out with individuals from polyploid populations but the results showed that there is a tendency towards self-fertilisation. The percentages ranged from 13 per cent to 30 per cent for cross-fertilised and from 0 per cent to 34 per cent for selfed flowers. It would appear, therefore, that though the polyploid plants may be self-pollinated, they may as readily be cross-fertilised. These remarks can only apply to the sea-level polyploids, however, since no plants were available from the much less dense hexaploid populations of the altitudinal ranges on account of the late maturing of the seed.

There is no marked colonial arrangement of the polyploid populations in the area. The octoploid population at sea-level where polyploids are frequent is partially isolated from the hexaploid populations but in close proximity to them. The occurrence of the hexaploids appears much more erratic on the gradients than is that of the diploids but field observations suggest that this is due to the relatively much reduced density of the former and not to any actual discontinuity of distribution. The size and general appearance of the hexaploid at the higher altitudes is so similar to that of the diploids that it is difficult to distinguish between them without close examination. Its presence to the altitudinal limits shown in Figure IV is, however, unquestionable though no data are available for the hexaploid above 1800 feet for reasons already given.

3. <u>Intergroup fertility</u> (diploid-hexaploid). Some trials were attempted but no reliable data have been obtained for intergroup fertility for two reasons. In the first place, several of the polyploids failed to flower when cultivated in pots entirely under greenhouse conditions and again, the polyploids flower at least one week later than the diploids. However, considering the extreme rarity of the tetraploid sexual plants in the wild even at sea-level where all groups may be found within crossing distance of each other, it may be concluded that interbreeding is very uncommon if it takes place at all. It seems more likely that the solitary specimen of a sexual tetraploid which was found is a representative of the tetraploid race within the setaceous-leaved sexual fescues rather than a hybrid between the diploid and the hexaploid, and even in the absence of experimental proof, it does not seem unreasonable to deduce that the two groups of sexual fescues, the setaceous-leaved and the broad-leaved represent independent genetic entities.

Jenkin (1934) made reciprocal crosses between Festuca rubra L. (2n = 42 or 56) and F. ovina L. (2n = 28) but only with F.ovina as the female parent was any germinable seed produced and then only 3.1 per cent. The poor seedlings failed to survive. It would seem, therefore, that there are intrinsic barriers to successful crossing between the species as well as the seasonal isolation although the latter factor alone could be sufficient to keep the species distinct. Anderson and Sax (1936) are of the opinion that Tradescantia hirsuticaulis, because of its early and short blooming season, gives very little evidence of crossing with T. virginiana, T. canaliculata and T. subaspera var. montana although it is occasionally associated with them.

In regard to the setaceous-leaved sexual fescues, it is interesting to note that no triploids were found in the wild. It is true that only one tetraploid was collected but it seems to suggest that tetraploids are present within the area and in that case, triploid hybrids might have been expected. Olssen and Rufelt (1948) found that in experiments with Sinapsis alba the triploid embryos formed as a result of interbreeding between diploid and tetraploid strains are very frequently abortive and they also quote Muntzing (1948) who found a similar state of affairs where diploid and tetraploid varieties of rye are grown together in yield trials. It may be, therefore, that crossing does take place between the diploid and tetraploid races of the setaceous-leaved sexual fescue but that the embryos are frequently abortive, which would account for their absence in the collection and possible rarity under natural conditions. This question also requires further clarification.

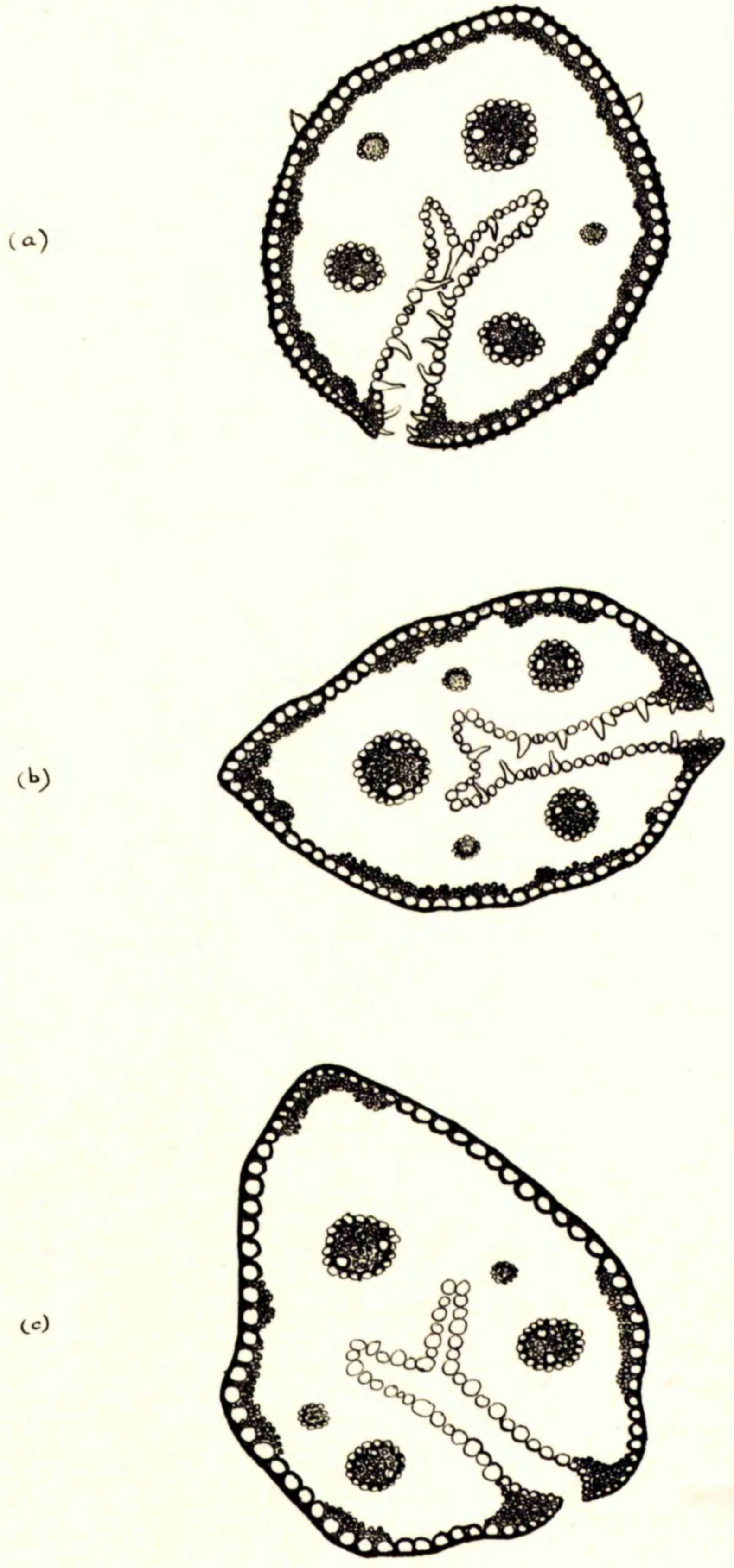
V. THE RELATIONSHIP BETWEEN CHROMOSOME RACES AND TAXONOMIC GROUPS.

Morphologically the sexual fescues from the Ben Cruachan area are divided into two main types, one with setaceous or capillary leaves and a split leaf sheath and the other, a larger plant with leaves flattened on the flowering stem at least and an entire leaf sheath. The former may readily be identified as Festuca ovina L. sensu ampliss. In recording 2n = 28 for a strain of F. ovina L. from Wales, Jenkin(1934) mentioned that the capillata form of F. ovina L. (= F. tenuifolia Sibth.) is said to be diploid. This would suggest that the Ben Cruachan setaceous-leaved sexual fescues may be identified as F. tenuifolia Sibth. but a closer examination of external characteristics renders such an equation of taxonomic group with chromosome number difficult to maintain (Table III). According to Howarth (1924), in F. tenuifolia the fertile glumes are typically awnless but may have awns up to 1 mm. in length whilst in F. ovina L. sens. strict, the fertile glumes have awns of 1 mm. or more in length. The large majority of the plants collected in the wild and of those grown from seed had muticous or mucronulate fertile glumes but some had small awns up to 0.6 mm. in length. They could, therefore, from the point of view of this character be grouped under F. tenuifolia but measurements of lamina breadth taken from plants grown from seed agree more nearly with those recorded for F. ovina L. sens. strict.. Again transverse sections of the radical laminae ranged from that described for F. tenuifolia to that for F. ovina sens. strict. with intermediate combinations of the two types. The lamina breadth of the

TABLE III.

Comparison between Festuca ovina L. species (Howarth) and Garden fescues.

	Festuca capillata Lam.	Festuca ovina L. sensu. strict.	Festuca supina Schur.	Garden fescues.
Laminae Breadth	0.3-0.5 mm.	0.3-0.65 mm.	0.5-0.6 mm.	0.4-0.9 mm.
Transverse Section	Continuous ring of sclerenchyma, rounded almost circular not keeled	Broken ring of sclerenchyma more keeled than F.capillata.	Continuous layer of sclerenchyma, 1-costate, eliptical, not keeled	combinations of characters.
Vascular bundles	5 - (7 rare)	5 - 7	7 - (5)	4 - 7
Culm Length	-40 cm.	-60 or 70 cm.	12-30 cm.	31-74 cm.
below panicle	angular or almost rounded, scaberulous or almost smooth or puberulous	rather angular scabrous or puberulous, rarely smooth	four-angled and scaberulous or puberulous	Various.
number of nodes	2	2	2	2
Panicle length	2-8 cm. compact	2-12 cm. more open	-4 cm. compact	4-13 cm. compact-open.
Sikelet length	3.0-5.5 mm.	4.8-8.0 mm.	6.8 (-9.5) mm.	2-9 mm.
Flowers number	3 - 8	3 - 8	4	2 - 8
Awn length	muticous (blunt) or shortly mucronulate	mucronate or -1 mm.	short	0-0.6 mm.
Sheath	smooth or feebly rough.	rather rough or quite smooth, sometimes hairy.	smooth	
	open to base	open to base	closed in lower third or quarter.	mainly open to base.



FESTUCA OVINA.

TRANSVERSE SECTIONS OF LEAVES FROM POPULATIONS

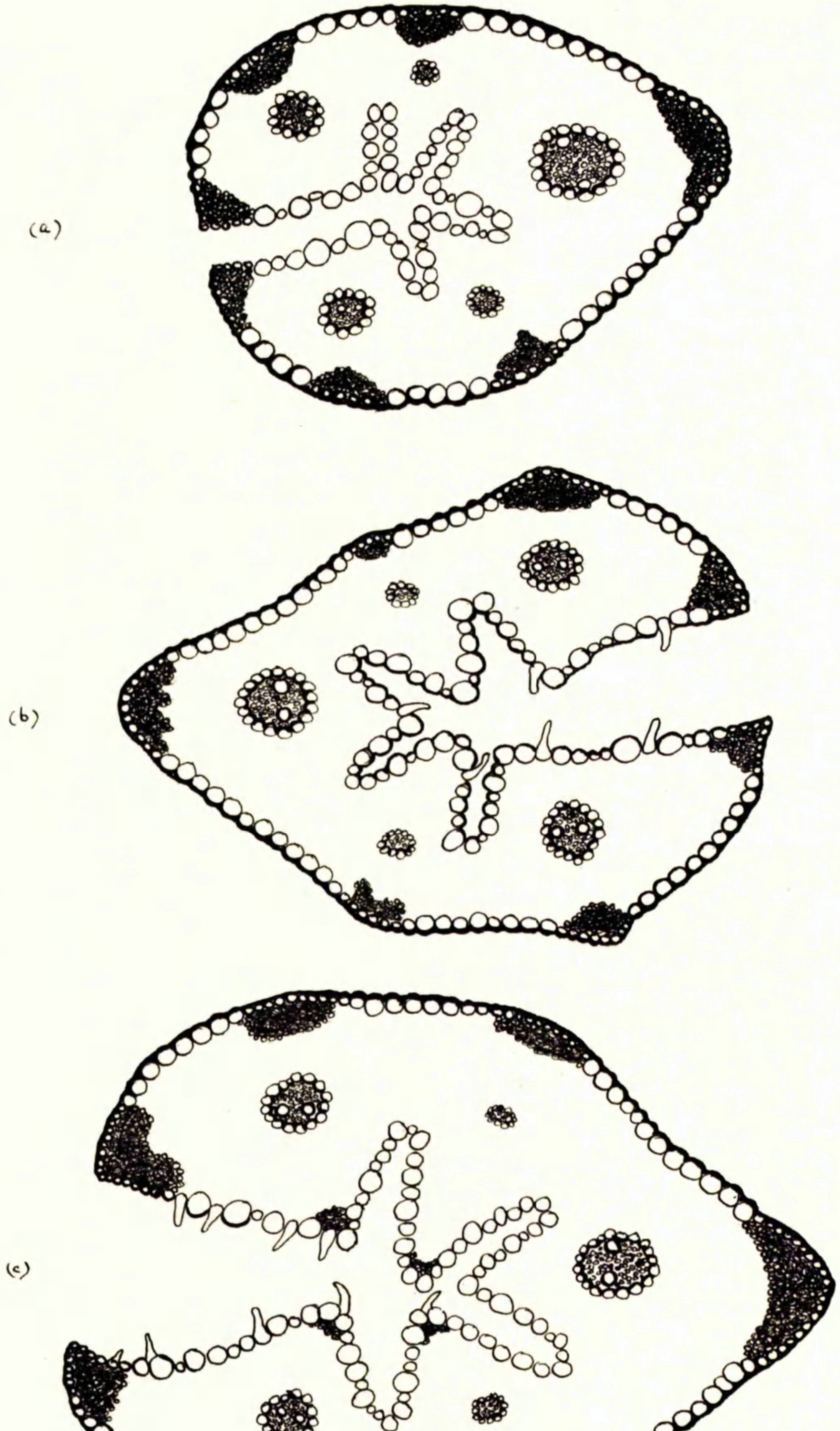
(a) 10 (TAYNUILT A, 0'), (b) 25 (TAYNUILT B, 2000'), (c) 40 (DALMALLY, 1500')

×90.

tetraploid from sea level is not significantly greater than that of the diploids and the fertile glumes are typically very shortly awned. Further, an examination without measurements, of the material collected in England and Wales and already mentioned in Table II, showed that plants with 2n = 28 chromsomes frequently had the characteristics of F. tenuifolia Sibth. There seems to be no evidence, therefore, for supposing that F. ovina L. sens. strict. is tetraploid whilst F. tenuifolia is diploid. The two chromosome races undoubtedly exist within F. ovina L. sens. ampliss but the data at present available suggest that the chromosome groups cut across the recognised taxonomic groups. As far as the specimens from Ben Cruachan are concerned, it would seem that in general they resemble F. tenuifolia most closely but also display F. ovina characteristics and may be considered as intermediates.

In view of the morphological difficulties encountered, a representative number of pressed specimens of these sexual fescues were sent to Dr. W. O. Howarth with his permission and he very kindly gave his opinion on their identification. Some he identified definitely as F. ovina L. others as F. tenuifolia Sibth. while others he thought were nearer to one than the other but appearing to be intermediates, were very hard to place.

The type with some flattened leaves and an entire leaf sheath may be broadly identified as F. rubra L. sensu. ampliss. Chromosome numbers of 2n = 42 and 2n = 56 have been quoted by Jenkin (1934) from data provided by Dr. Sethi for F. rubra material found in Great Britain and Lewitsky and Kuzmina (1927)



FESTUCA RUBRA

TRANSVERSE SECTIONS OF LEAVES FROM SEA-LEVEL POPULATIONS.

(a) 29 ($2n = 42$) (b) 30 ($2n = 42$) (c) 9 ($2n = 56$). × 90.

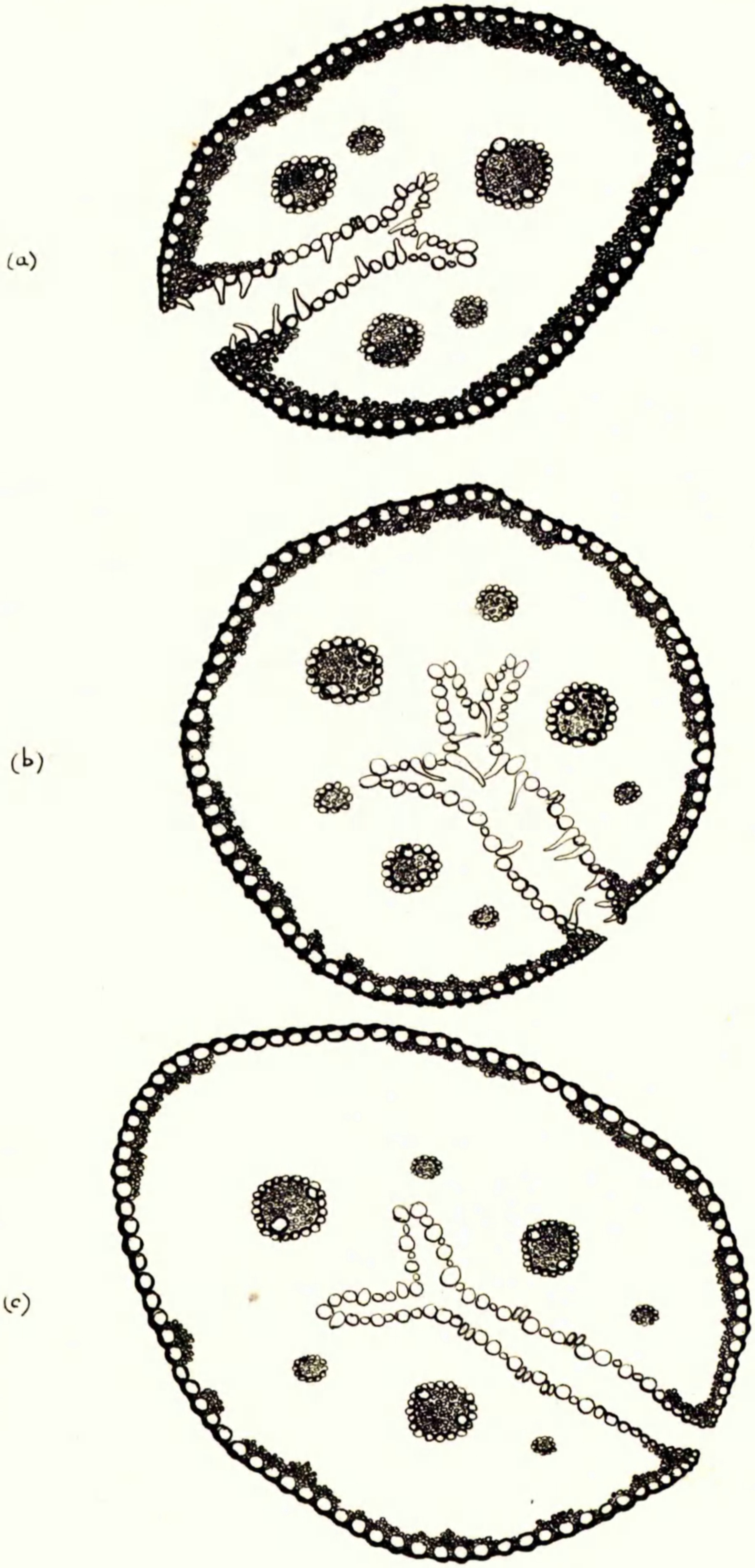
reported 2n = 56 for *F. rubra* subsp. *eurubra* var. *genuina* Hack whilst Stählin (1929) gave 2n = 42 for *F. rubra* subsp. *eurubra* var. *genuina* subvar. *vulgaris* (= *F. rubra* L. *vulgaris*). Maude gives 2n = 42 (1939) for *F. fallax* so that there is no possibility of identifying the hexaploid chromosome number with *F. rubra* alone and it would seem that again chromosome races exist within the group sens. ampliss. without any connection with any one taxonomic grouping. With reference to the Ben Cruachan *F. rubra* types, no distinguishing differences may be detected between the hexaploid and the octoploid plants and on the other hand, variation is present, making some known hexaploids nearer to the octoploids than to other hexaploids. A further source of difficulty is present in the form of the root stock. Only some of the plants are decidedly stoloniferous whilst others have an almost entirely tufted habit of root-stock growth which is characteristic of *F. fallax*, but on good authority it has been decided to refer to this group in general as *F. rubra*, (Table IV).

The taxonomic classification of the viviparous fescues presents a little difficulty. The chromosome number does not provide any clue since different races ranging from 2n = 21 to 2n = 56 (Turesson 1930, 1931; Flovik 1938, 1940) have been reported though only 2n = 28 has been found in Britain. Turesson came to the conclusion (1926) that all the viviparous fescues should be grouped under *F. ovina* subspecies *vivipara* and that further subdivisions should be termed "apomict" or "amphiapomict" according to their degree of vivipary. While

TABLE IV.

Comparison between varieties of Festuca rubra L. genuina (Howarth, 1923) and Garden fescues.

	F. rubra L. vulgaris	F. rubra L. grandiflora	F. rubra L. tenuifolia	F. rubra L. glaucescens	F.rubra L. dumetorum	F. rubra L. planifolia	F. rubra L. juncea	F. rubra L. arenaria	Garden fescues
Laminae radical	setaceous 0.5-1.0mm.	0.8-1.0mm. or more	0.6-0.8mm.	0.5-0.8mm	0.7-1.1mm.	2-3 mm.	0.8-1.2mm.	0.9 or more	0.6-2.4mm.
Panicle length	-18 cm. compact-loose	10-18 cm. erect, open	6-9 cm. compact, open at anthesis	2.5-7.0 cm. compact, open at anthesis	11-14 cm. ?	large loose, multi-spiculate	5-11 cm. large, somewhat lax.	7-13 cm. ?	6-21 cm. erect, open.
Spikelet Length	5-8-10mm.	10-13 mm.	-12 mm.	6-14 mm.	8-11 mm.	9-10 mm.	9-10 mm.	9-13 mm.	5-15 mm.
Flowers number	5-7	5-8	-8	4-7	4-7	many	?	4-8	2-9
Awn length	-2 mm.	-3 mm.	mucronate -aristate	?	?	short	short	aristate	0.1-2.0mm.



FESTUCA VIVIPARA.

TRANSVERSE SECTIONS OF LEAVES FROM POPULATIONS

(a) 6 (TAYNUILT A, 1200'), (b) 27 (TAYNUILT B, 1450'), (c) 16 (TAYNUILT B 3689')

×90.

accepting the main plan of Turesson's key to classification, Wilmott (1945) decided that for ease of reference it would be better to regard the viviparous fescues as a separate species, F. vivipara, and further subdivisions would consequently be given the rank of subspecies. In this he agrees with a number of writers including particularly Scholander (1934) who based his conclusion on the belief that there could be no continuous transition from F."ovina" (presumably sensuampliss) to F. "vivipara" as no awns are present in F. vivipara whatever the degree of vivipary. This would scarcely seem to be a valid argument since F. ovina subspecies capillata is distinguished from F. ovina sensu strict. in having no awns, and in some cases awns have been found on viviparous types and are actually used in the classification of types or subspecies (Wilmott).

Regarding the British viviparous fescues alone, the difference between them and the sexual forms is very striking in the wild as far as reproduction is concerned and it would seem justifiable to rank them as a separate species but taking the area of distribution as a whole, it has to be remembered that there are also semi-viviparous forms and though much work remains to be done, it seems possible that in them may be found the explanation of much which is at present puzzling. In the present state of knowledge and particularly in view of the different senses in which the terms are used, it does not seem to matter whether for ease of reference, the viviparous forms are given specific or subspecific rank as long as it is remembered that different chromosome races exist, the relationship between which remains to be experimentally determined. It is of interest to note that Gustafsson (1947, p.267) is of the opinion that "there is no special need to specify and name the individual apomicts more than the biotypes of a sexual species."

Wilmott adapted Turesson's key to cover the various forms which were found on the Uig peninsula and this key has been used in the present work.

KEY TO THE IDENTIFICATION OF VIVIPAROUS FESCUES
(pp. 52 and 53 Flora of Uig)

I. Glumes I and II not visibly metamorphosed. (II: with glume II in apical spikelets metamorphosed, 8-12 mm. long, sometimes longer, includes a single Norwegian subspecies.)

A. The three or four lowest glumes, at least on basal spikelets, not at all or slightly metamorphosed: glume IV not twice as long as glume II.

/ Six to eight glumes at the base of the spikelet unmetamorphosed. ssp. uigensis mihi.

// Four glumes at the base of the spikelet not at all or slightly metamorphosed.

1. Glume II, as the rest of the lower glumes, short hairy.

a. Glume I in the middle and apical spikelets with a short point (c. .5 mm. long. (b: ... with a long drawn out point - includes one subspecies rather widespread in northern Scandinavia.)

* Ripe bulbils long and narrow, clustered in a thick bushy panicle, and with rather strong adventitious root formation.
ssp. faeroensis (Turess.) comb. nov.

** Bulbils medium sized (to 35 mm. long), in a moderately lax panicle, and with weak adventitious root formation.
ssp. jemtlandica (Turess.) comb. nov.

2. Glume II glabrous, like the rest of the lower glumes, or at most at the apex with small sparse stiff hairs.

a./

a. Glume I in the middle and apical spikelets with a short point. (b: ..., with a long drawn out point - one subspecies widespread in Torne Lappmark).

* Ripe bulbils long and narrow, clustered into a thick bushy panicle, with strong adventitious root formation.
ssp. vaagensis (Turess.)
comb. nov.

** Bulbils of medium size, in moderately lax panicles, and with weak adventitious root formation.

/ Glume II gradually narrowed to the apex. (//: ... with truncate apex - one Swedish subspecies restricted to Areskutan.)
ssp. norvegica (Turess.)
comb. nov.

B. Glumes III and IV in middle and basal spikelets metamorphosed. Glume IV twice the length of glume II.

(This section includes nine Scandinavian subspecies and two Scottish, but no specimens collected in 1939 in the Outer Hebrides appear to belong here. As the two Scottish subspecies have not yet been introduced into British literature, I add their description, extracted from Turesson's key:

ssp. killinensis (Turess.) comb. nov. Panicle broad and bushy; culm weak, soon inclined; laminae of bulbils without markedly raised nerves - Glume II glabrous, like the rest of the glumes, or at most with small sparse stiff hairs near the tip - Glume II in middle and basal spikelets 4-5 mm. long, often pure green like the rest of the lower glumes - Glume I in middle and apical spikelets with only a short point. - Perthshire: Killin.

ssp. scotica (Turess.) comb. nov. Panicle broad and bushy; culm weak, soon inclined; laminae of bulbils without markedly raised nerves - Glume II short hairy, like the rest of the lower glumes, c. 1 mm. broad, panicle not secund - Bulbils medium sized (to 35 mm. long) - Glume I in middle and apical spikelets with a short point. - More widely spread in Scotland.

The identification of the viviparous fescues by means of the key was rendered dubious particularly by the length of the awn in middle and apical spikelets and by the fact that Glume IV was very frequently twice the length of Glume I in basal spikelets (Table V.)

TABLE V.

Comparison of *Festuca vivipara* varieties with garden fescues.

	F. vivipara faeroensis	F. vivipara vigensis	F. vivipara norvegica	F. vivipara scotica	Garden viviparous fescues
Number of glumes wholly or partially metamorphosed (basal spikelets)	4	6-8	4	2	2-7
Glume I. (middle & apical spikelets) Awn length	c.0.5 mm.	c. 1 mm.	c.0.5 mm.	c.0.5 mm.	Blunt - 0.5 mm.
Glume II. Hairiness	short hairy	subglabrous	glabrous or with small sparse stiff hairs at apex.	short hairy	Glabrous - spreading hairs.
Glume IV. Length	not twice as long as Glume II.	not twice as long as Glume II.	not twice as long as Glume II.	twice as long as Glume II	Both.
Ripe bulbils	long and narrow in thick bushy panicle	?	medium size rather lax panicles	medium size	long and straight and curled leaves.
Root formation	strong	?	weak	?	mainly strong.

One or two plants might be identified under Turesson's description of *F. ovina vivipara* aapm. *scotica* but the majority came somewhat doubtfully under aapm. *faeroensis* or aapm. *uigensis*. Almost without exception, however, at least one character would fail to fit into the description although taken as a whole, a plant might agree more with one than with another.

It will be seen that the range of variation among the garden fescues is wide and since it exists not only between, but also within populations and even in individual plants, it seems reasonable to conclude that any taxonomic varieties based on such variable attributes must be regarded with some suspicion. A similar conclusion must be drawn from the brief discussions on the *F. ovina* and *F. rubra* types thus giving further proof of the value of an examination of areal and ecological distribution of variation within the genetically independent units before attempting to delimit infra-specific taxonomic categories.

In cases where it is necessary to refer to groups in taxonomic terms in the following discussion, the undernoted nomenclature will be adopted.

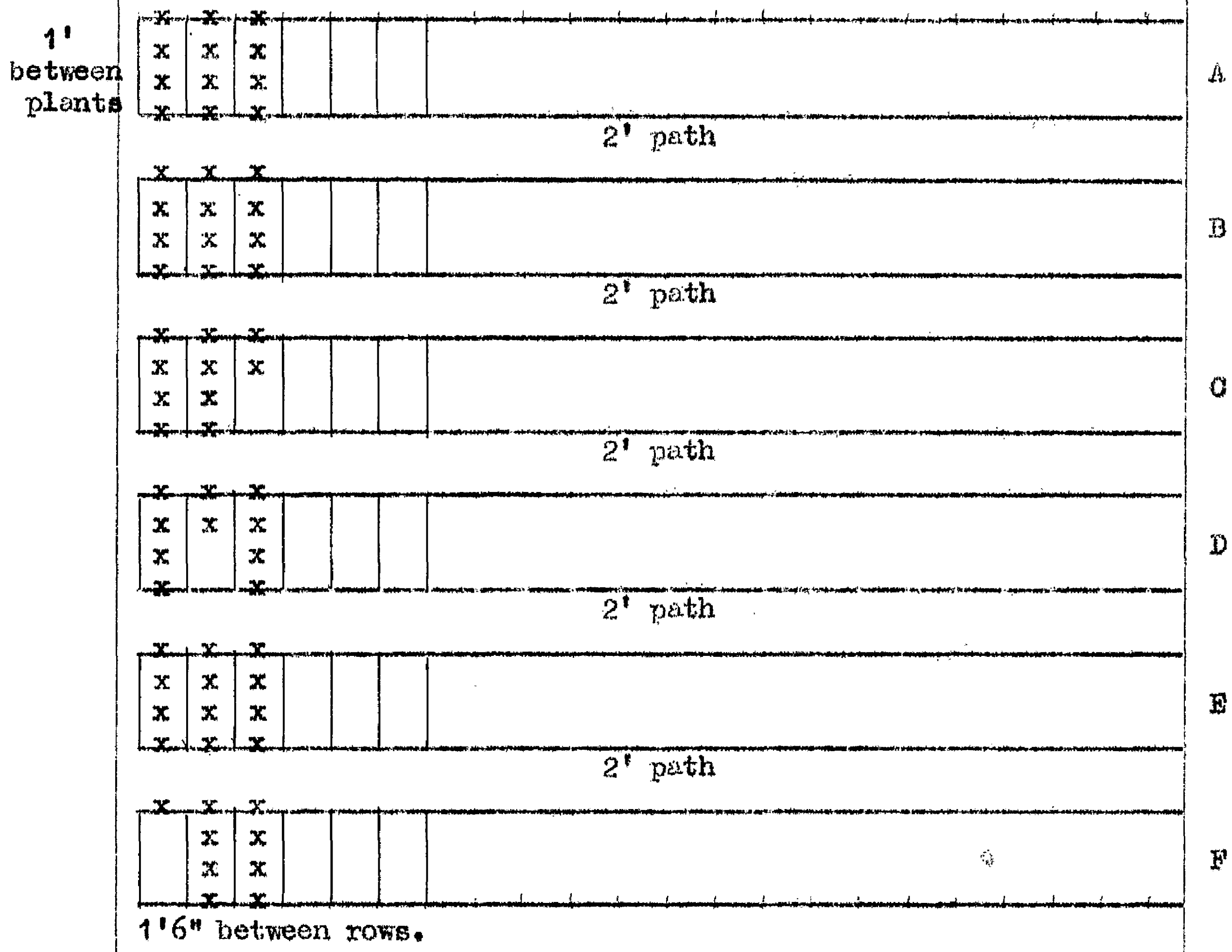
Sexual setaceous-leaved type with split leaf sheath, *F. ovina*
Viviparous setaceous-leaved type with split leaf sheath, *F. vivipara*.

Sexual types with leaves flattened on flowering stem at least and entire leaf sheath, *F. rubra*

VI. EXAMINATION OF THE MATERIAL.

It was necessary for the purposes of the present investigation to examine living material growing under conditions of a uniform environment, and, therefore, seed and bulbils were collected from the gradient stations in the late summer of 1944 and sown in weed-free soil. Scarcity of seed and the failure of some bulbils to take root reduced the number of plants available from some stations to less than the desired twenty-four but in no case were less than six plants used. They were transplanted in the experimental garden in replicated plots as illustrated in Figure V.

Figure V.
Arrangement of Blocks.

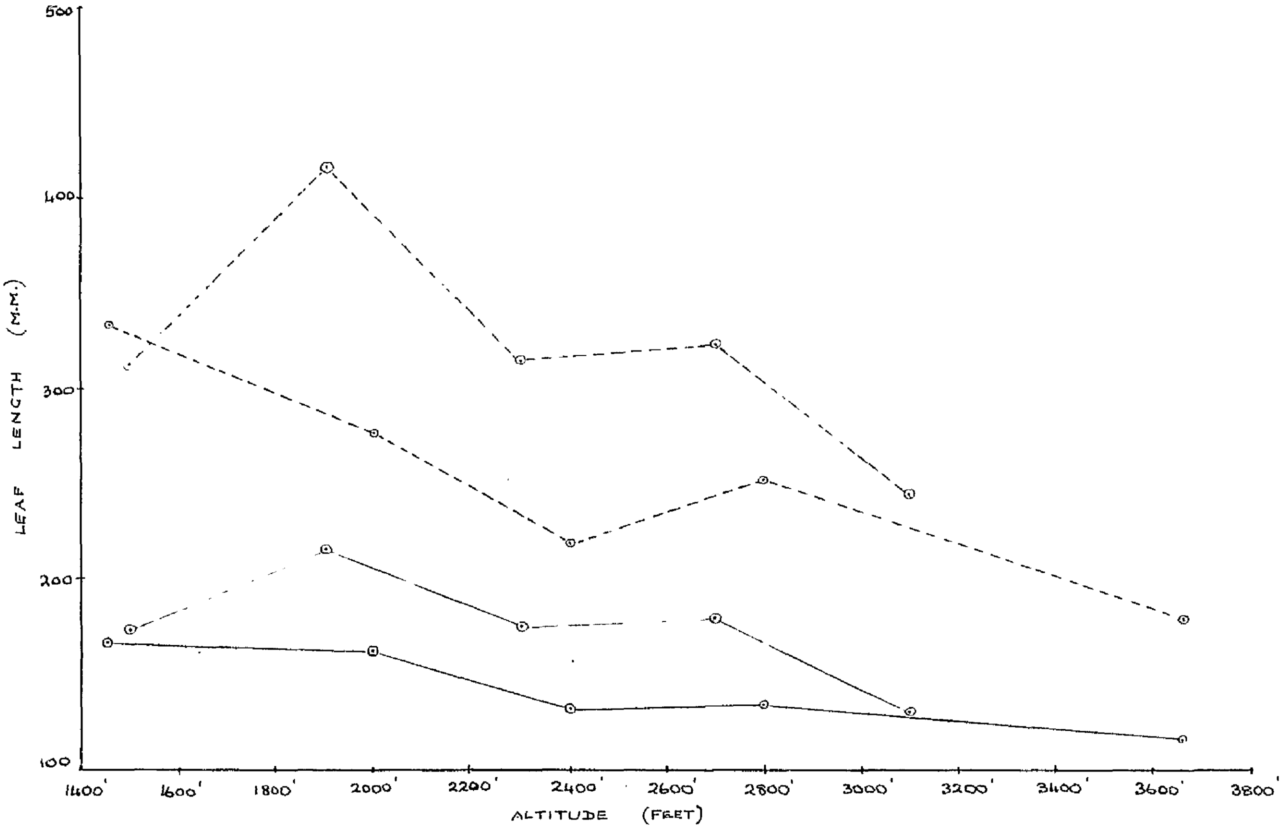


F. vivipara plants were planted in a separate layout from F. ovina and F. rubra plants but the same method of replicated plots was employed. A second collection of seed and bulbils was made in the late summer of 1945 and the same procedure was followed. It had been observed, however, that the spacing was rather limiting for the large F. rubra plants and it was increased for all the sexual plants to 2 feet all round but left unchanged for the F. vivipara plants. Each lot was retained for examination over a period of two years.

Since it was considered impossible to obtain for each plant a mean value from a number of observations, the principle of homotypic measurement which was discussed and adopted by Gregor et al. (1936), was used. The measurement of Leaf Length and Leaf Breadth provide an example. During the first year of growth when the plants were purely vegetative, the longest radical leaf was taken from each plant and the length and breadth measured. This was repeated twice during the season and the graphs of the means for each population proved to be sufficiently alike in direction and curve for the three dates to justify this method of measurement and to establish the comparative accuracy of the results (Graphs I and II).

The data were examined statistically, use being made in particular of the Correlation coefficient and the Analysis of Variance. Several difficulties arose in the method of calculation for the Analysis of Variance because of the absence of plants in a few plots owing to deaths and other causes and to unequal numbers of plants occurring in different plots of the same population. Dr. R. A. Robb of the Mathematics Department

GRAPH I

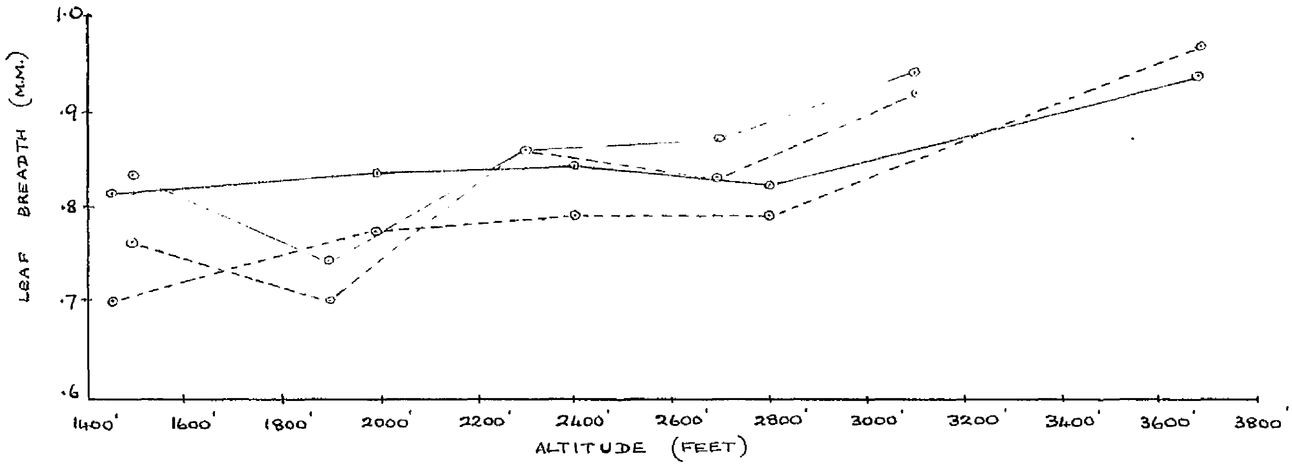


LEAF LENGTH MEASUREMENTS FOR JUNE AND SEPTEMBER 1945.

TAYNUILT B JUNE ———— DALMALLY JUNE ————

SEPTEMBER - - - - - - - - SEPTEMBER - - - - - - -

GRAPH II



LEAF BREADTH MEASUREMENTS FOR JUNE AND SEPTEMBER 1945.

TAYNUILT B JUNE ———— DALMALLY JUNE ————

SEPTEMBER - - - - - - - SEPTEMBER - - - - - - -

of Glasgow University gave very generous guidance on the modifications necessary and a detailed note of the techniques used is given in the Appendix.

A simple Analysis of Variance giving only the significance of the variation between populations had to be used for the sexual 1946 plants as, owing to a shortage of growing space, the complete layout could not be accommodated as a single unit. The two portions were planted in adjacent areas separated by a 4 feet path and since the homogeneity of the conditions had been demonstrated in previous experiments, it was considered justifiable to use the results of the Analysis of Variance calculations with the exception of the significance between Blocks.

Most emphasis must be placed upon the results for the 1945 plants since although in some cases a larger number of altitudinal populations were available for examination in 1946, a wider range of measurements was obtained for the 1945 plants. It is for this reason that Correlation Coefficients between altitude and the various attributes have, generally speaking, been calculated for 1945 plants only. On the same principle, Divergence Indices (Ginsberg 1938) have been calculated for the 1945 plants.

The material collected from the various gradients and used in the garden experiments is summarised in Tables VI and VII.

TABLE VI.

Populations on Taynuilt Gradients A and B
F. ovina and F. vivipara.

Station	Altitude	F. ovina		F. vivipara	
		1945	1946	1945	1946
	feet				
30	0	x	x	x	x
8	0		x	x	
1	200		x		x
2	400		x		x
3	600		x		x
4	800		x		x
5	1000	x	x		x
6	1200	x	x		x
7	1400	x	x		x
14	1600	x			x
27	1450	x	x	x	x
26	1800				x
25	2000	x	x	x	x
24	2200	x	x		x
23	2400	x	x	x	x
22	2600	x	x		x
21	2800			x	x
20	3000				x
19	3200				x
18	3400				x
17	3600				x
16	3689			x	x

TABLE VI(a).

Populations in Taynuilt Gradients A and B
F. rubra

Station	Altitude	1945	1946
	feet		
8	0	x	x
9	0	x	x
10a	0		x
29	0	x	x
30a	0	x	x
1a	200	x	x
2a	400		x
27	1450	x	x
26	1800	x	

TABLE VII.

Populations on Dalmally Gradient
F. ovina and F. vivipara.

Station	Altitude	F. ovina		F.vivipara	
		1945	1946	1945	1946
	feet				
63	400			x	x
35	500				x
36	700				x
37	900	x	x		x
38	1100			x	x
39	1300				x
40	1500	x		x	x
41	1700				x
42	1900			x	x
43	2100				x
44	2300			x	x
46	2700			x	
62	3100			x	

TABLE VII(a)

Populations on Dalmally Gradient
F. rubra

Station	Altitude	1945	1946
	feet		
63	400	x	x
35	500	x	x
36	700	x	x
37a	900	x	
39	1300	x	

Character Measurements

The longest radical leaf from each plant was measured in millimetres for Leaf Length and Leaf Breadth as has already been explained. Plant Height was taken as the height in millimetres of the longest flowering stem and the panicle on this stem provided the basal spikelet on the lowest rachis for the measurement of Spikelet Length and Awn Length. These two measurements were made using a X4 eyepiece with a micrometer scale in conjunction with the horizontal Vernier scale on the mechanical stage of a microscope. The Awn Length figure obtained by this method has not been converted into millimetres for the purposes of the calculations. The number of flowers in the basal spikelet and the number of spikelets on the panicle were counted at the same time.

Potential Fertility was calculated for each plant from the figures for the number of flowering stems, the number of spikelets per panicle and the number of flowers per spikelet. The Actual Fertility was similarly calculated from the number of heavy seed per panicle and the number of flowering stems.

At the end of the second year of growth all the 1945 plants were lifted, shaken as free of soil as possible, and individually weighed to give the total weight of stock and root growth. With the 1946 plants the stock part of the plant was skimmed from the roots with a sharp spade to give the weight of the upper part of the plant only.

The Habit of Growth ratio was calculated from measurements Flowering Stem Spread/Plant Height. The measurements were taken when the panicles had generally fully emerged from the sheath but were not yet flowering. Plant Height in this connection is the height of the tallest part of the plant above the ground and does not necessarily agree with the height of the longest flowering stem.

This method proved to be invalid for the sexual polyploids which have almost without exception an erect habit of growth, as far as the flowering stems are concerned at any rate. The spread in the case of these plants includes the degree to which stolons have been developed as well as the laxity or rigidity of the flowering stem. The latter point renders the ratio less accurate also for the plants to which it has been applied since it does not permit of a distinction between a genuinely prostrate plant and one which has a very lax flowering stem, but at least it is the only complicating factor.

The Time of Flowering was recorded, according to the following scale, at weekly intervals from the first appearance of the panicles until flowering had ceased. The recording for 19th June has been used for the statistical calculations.

Flowering grade. Sexual Plants.

1. Apex of panicle appearing from sheath.
2. Panicle clear of sheath.

2a. 50 per cent of panicles clear of sheath.

3. Panicle expanding.
4. Plants starting to flower (up to 12 per cent panicles flowering).
5. Plants flowering sparsely (up to 25 per cent panicles flowering).
6. Plants flowering freely (up to 50 per cent panicles flowering).

7. Plants flowering abundantly (up to 75 per cent panicles flowering).
8. Panicles not closing but not still flowering.
9. Flowering completed but panicles not all closed.
10. Panicles at least 50 per cent closed.
11. Panicles all closed.
12. Some panicles yellowing.

Flowering grade. Viviparous Plants.

1. Apex of panicle appearing from sheath.
2. Panicle clear of sheath.
3. 50 per cent clear of sheath but none so far expanded.
4. Earliest panicle expanded.
5. Panicles 12 per cent expanded.
6. Panicles 25 per cent expanded.
7. Panicles 50 per cent expanded.
8. Panicles 75 per cent expanded.
9. All expanded.
10. Panicles closing.
11. Panicles half closed.
12. Panicles closed.

In order to obtain a figure for the amount of seedling growth made by seeds from different altitudinal populations twenty seeds were weighed and planted in sand. The seedlings were removed as carefully as possible at the end of three months, all particles of sand washed from the roots and after a momentary pressure between sheets of absorbent paper, weighed quickly. The weight of the seed was subtracted from that for the seedling and the mean value calculated from the resulting figures was used as an estimate of seedling vigour. The experiment was repeated in the following year.

TABLE VIII.

1945. Diploid Series, F. ovina. Transect A.

		Leaf Length		Leaf Breadth		Plant Height		Plant Height		Habit of growth		Flowering Grade		Potential Fertility		Actual Fertility		Spikelet Length		Awn Length	
Population	Altitude feet	MV mm	DI	MV mm	DI	MV 100s of mm	DI	MV 100s of mm.	DI	MV ratio	DI	MV	DI	MV 100s of flowers	DI	MV 100s of seeds.	DI	MV mm	DI	MV units	DI
30	0	213		.5910		4.8		7.3		1.13		4.6		1034		545		4.9		5.8	
5	1000	249	61	.5671	63	5.6	72	6.8	55	1.33	63	5.3	52	1090	53	556	52	5.7	68	6.3	61
6	1200	* 209	62	.5805	64	5.3	72	7.6	56	1.01	63	5.4	57	904	61	502	54	5.4	61	7.6	60
7	1400	210	55	.6195	55	5.1	63	7.7	60	1.16	55	5.4	54	653	59	356	59	5.0	63	6.1	63
14	1600	** 271	73	.6120	58	5.1	57	9.1	58	1.09	52	6.4	64	729	59	371	53	5.6	65	6.1	62
Divergence Index between end points			69		59		58		60		54		69		66		63		69		77
Significance between end points		**										**									
Correlation coefficient Character: altitude												+.132									

* significant at ·05 point
** significant at ·01 point } (F. test Snedecor (1946) pp. 218 - 226)

TABLE IX./

TABLE IX.

1945 Diploid Sexual, _F. ovina._ Taynuilt B.

Population	Altitude	Leaf Length		Leaf Breadth		Plant Height		Plant Weight		Habit of growth		Flowering Grade		Potential Fertility		Actual Fertility		Spikelet Length		Awn Length	
	feet	Mean Value mm.	Divergence Index.	MV mm	DI	MV 100s of mm.	DI	MV lb.	DI	MV Ratio	DI	MV	DI	MV 100s of flowers	DI	MV 100s of seeds	DI	MV mm	DI	MV units	DI
27	1450	298		.6213		6.3		7.8		1.01		5.5		924		535		5.1		7.4	
25	2000	** 195	71	.5986	65	** 5.2	77	8.6	72	1.25	51	4.2	53	716	69	506	57	4.9	62	7.3	65
24	2200	202	62	* .6648	62	5.3	63	8.4	61	1.11	59	6.8	57	600	59	354	73	5.4	66	7.2	66
23	2400	186	54	.6657	59	** 4.6	73	8.7	65	1.18	56	6.6	52	438	60	278	58	5.4	54	11.0	65
22	2600	* 148	67	.6152	63	4.2	68	9.4	64	** 1.44	62	6.1	53	354	61	226	55	5.1	63	8.4	63
Divergence Index between end points			95		86		89		72		81		57		75		77		54		54
Significance between end points		xx				xx				xx				xx		x					
Correlation coefficient Character: altitude		-.604 xx		+.037		-.224 x		+.103		+.186		+.197 x		-.437 xx		-.400 xx					

* SIGNIFICANT AT ·05 POINT

** SIGNIFICANT AT ·01 POINT

TABLE X.

1946 Diploid Sexual, F. ovina. Taynuilt A.

Pop.	Alt. feet.	Mean Character Values			
		LL	LB	PW	Fl.G.
		mm	mm	oz.	19/6
10	0	229	.6700	-	4.8
30	0	211	.5935	15.6	* 3.1
1	200	* 230	.6200	* 20.4	3.8
2	400	236	.6161	19.0	3.9
3	600	** 267	.6132	16.5	3.9
4	800	** 245	.5945	20.2	4.1
5	1000	** 296	.6400	** 15.0	4.9
6	1200	** 273	.6329	18.6	* 6.0
7	1400	276	.6170	19.0	* 5.0
Significance between end points		xx			
Correlation coefficient char:alt.					+.470 xx

TABLE XI.

1946 Diploid Sexual, F. ovina. Taynuilt B.

Pop.	Alt. feet.	Mean Character Values.			
		LL	LB	PW	Fl.G.
		mm	mm	oz.	19/6
27	1450	270	.6540	17.6	5.2
25	2000	** 192	.6312	13.7	* 3.7
24	2200	** 259	.6100	17.3	* 5.5
23	2400	** 209	.6317	14.0	6.3
22	2600	187	.6683	13.0	4.8
Significance between end points		xx			

* SIGNIFICANT AT ·05 POINT.

** SIGNIFICANT AT ·01 POINT.

VII. ANALYSIS OF CHARACTERS.

(a) Diploid. F. ovina.

Since it is not possible to examine the total sum of the attributes, both morphological and biological, which compose the individuals of a population, it would be invalid to attempt to form a true picture of the relative position of that population by considering the total of measurable characters only and their distribution. The greatest amount of information will be derived from an examination of the distribution of variation of each available character for each gradient. The data shows that the variation is distributed in three different ways.

1. In the first case, the Analysis of Variance calculations establish that there is not, at any rate, a single uniform population extending over the whole gradient for the character in question yet no trend in any direction may be detected. This is illustrated by randomly varying mean values for Leaf Length on the Taynuilt A (1945 and 1946) (Tables VIII and X) gradient. The individual plant records do not vary to any great extent from the mean for each population and in some cases, the differences between populations are statistically significant.

An explanation of the random occurrence of variation on the lower slopes of Ben Cruachan may be found in the opportunities afforded for colonisation. Though, as has been previously mentioned, there is at no altitude any strongly marked colonial development, it must be admitted that on the lower slopes there is a greater diversity of vegetational types. F. ovina is forced to compete for living room with many other species, often of greater height though not necessarily more capable of survival, and there are greater possibilities for a less dense colonisation than occur on the upper levels. The effective size of the breeding population is undoubtedly smaller under the more scattered conditions and small-scale genetic drift is probably responsible at least to some extent for these random differentiations. Woodson (1947) found examples of population values in Asclepias tuberosa in U.S.A. which do not fit in with the general genocline which he traced and attributes these apparent discrepancies to genetic drift "in which the influence of natural selection is absent or negligible" since he found that the colonies involved were small and occur relatively infrequently owing to locally unfavourable conditions.

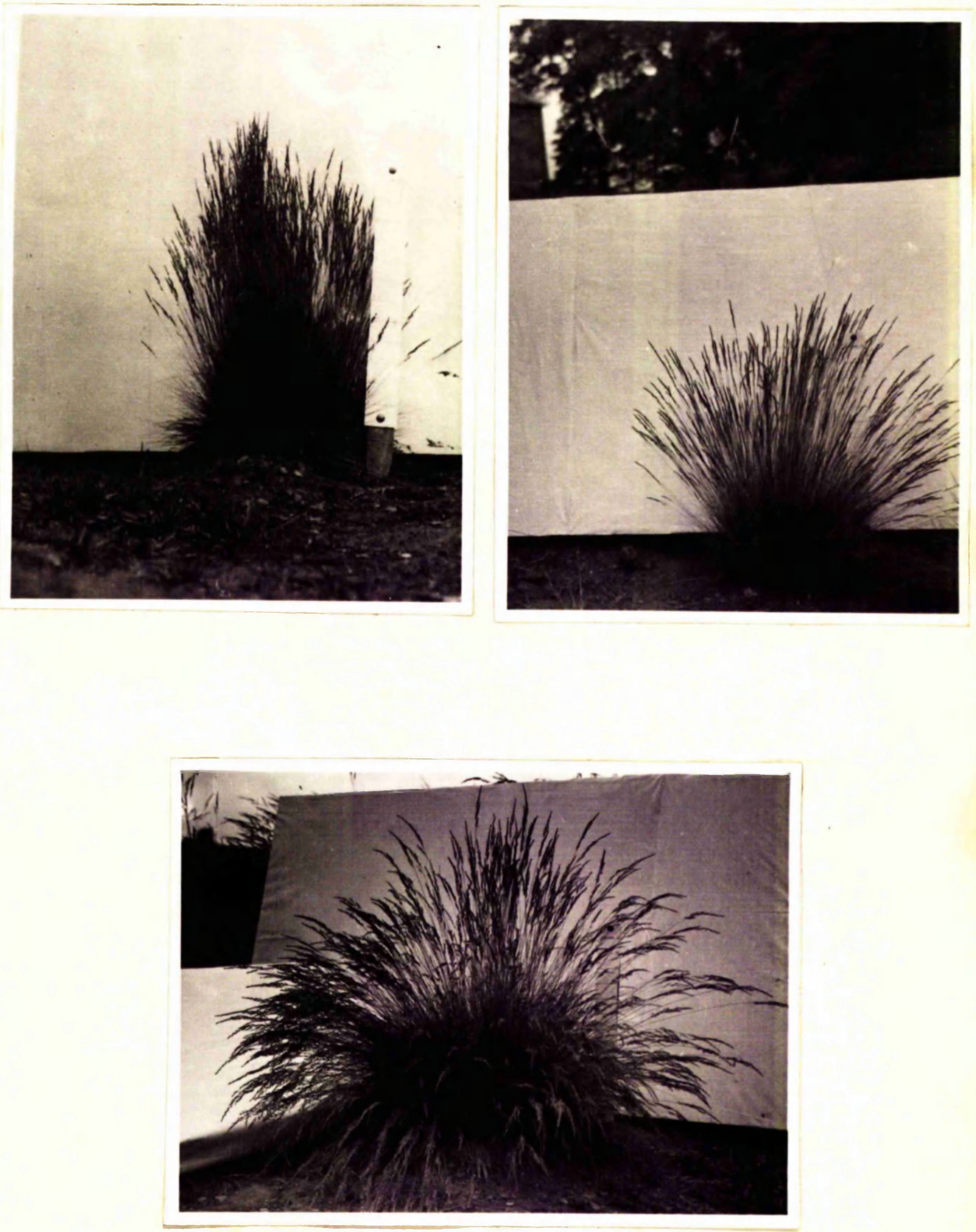
2. Secondly the absence of a single uniform population is proved by the Analysis of Variance calculations and a definite trend of change correlated with increase in altitude is present. An example of this type of variation distribution is shown markedly by Potential Fertility (1945) for the populations on the Taynuilt B gradient (Table IX) and is probably displayed by attributes which are most strongly influenced by the general environmental change.

3. In the third case, the Analysis of Variance calculations do not prove that any significant difference exists between populations and it can be seen from the individual plant records that the variability is as great within each population as it is between them. Spikelet Length and Awn Length for both Taynuilt

gradients, A and B (Tables VIII and IX) display this type of uniform distribution of variation. It seems likely that these attributes are of no selective value.

It may be remarked that whilst Leaf Length illustrates the first type of distribution of variation on Taynuilt A gradient (Tables VIII and X) it illustrates the second type on Taynuilt B gradient (Tables IX and XI). A possible explanation of the random occurrence of variation on Taynuilt A gradient has already been suggested but the more irregular climatic conditions must also be mentioned. In winter the upper slopes frequently have a covering of snow for considerable periods but the occurrence on the lower slopes is more erratic. The higher altitudes suffer a more severe climate and are at certain seasons of the year almost constantly wreathed in mist which often extends down to 1500 feet. On the whole it must be observed that though more exacting, the environmental conditions of the upper slopes are more uniform than they are at the lower levels.

It has already been established by the experiments on crossing that individuals from widely separated, and even from the extreme, populations are capable of interbreeding but there is ~~no~~ evidence from the distribution of variation that, in fact, the area of each gradient is not covered by a single population but by a series of partially independent populations. In particular, the large degree of divergence which is shown by the Divergence Indices for the extreme populations supports this view. Between adjacent populations, certainly, the degree of divergence may not be great but for the majority of the characters, it is sufficiently large



FESTUCA OVINA. EXAMPLES OF TYPES OF HABIT OF GROWTH.

to justify the claim that populations are at least to some degree individual. The amount of overlapping between neighbouring populations for any one character is always greater than the degree of divergence but the fact that it is present, makes it clear that there is a difference in the frequency with which the variate occurs in the populations in question. As has already been noted, trends associated with increase in height can be established but a glance at the Divergence Indices does not indicate that the amount of divergence between populations becomes either greater or less with increasing altitude.

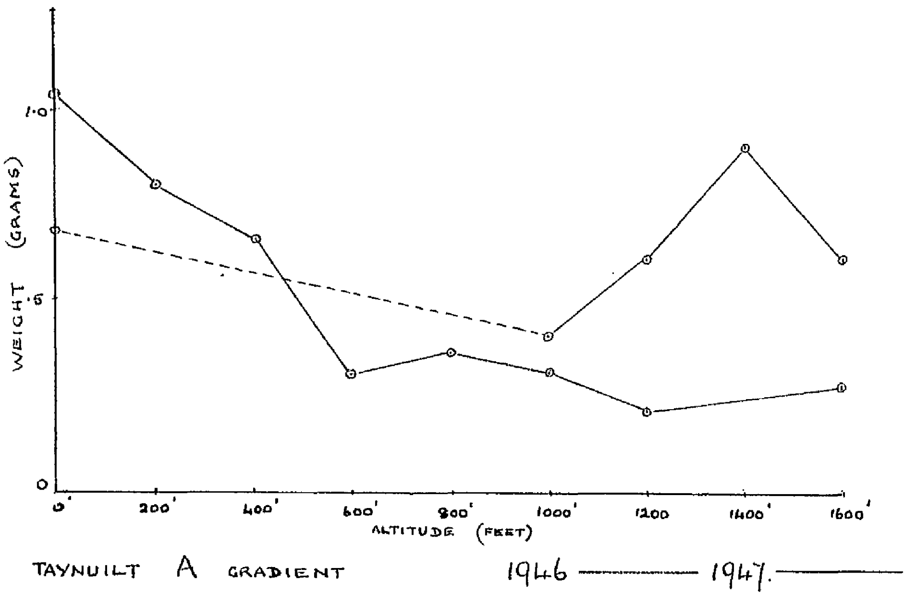
The presence of a significant difference between adjacent populations for one or even several attributes serves to emphasise the discontinuity which exists. If the populations were not to some extent independent, such significances would not occur but on the other hand, the absence of any evidence for significance in the case of some other characters points to the incompleteness of this independence. This may be illustrated by reference to populations 27 and 25 (Table IX); significant differences occur between these populations for both Leaf Length and Plant Height (1945) but for no other attributes.

The graph of the figures obtaining for seedling growth in 1946 is strikingly different from that for 1947.

Graph III/

GRAPH III.

Comparison of Diploid Seedling Growth
in 1946 and 1947.



The graph for 1946 shows a result which might be expected since there is a decrease in the amount of growth as altitude increases. The irregularities of the graph for 1947 are surprising but a possible explanation lies in the fact that whereas the seed used in 1946 was collected from the wild populations themselves, the seed used in 1947 was taken from the diploid plants growing in the garden. The opportunities for cross-fertilisation were much greater under the artificial conditions and it is very probable that some of the seed was the result of crossing between parents which are normally spatially isolated. The only reliable indication of seedling vigour must be obtained from the graph for 1946 which suggests at least that growth tends to proceed more slowly as altitude increases.

(b) Sexual polyploids, F. rubra.

(1) Taynuilt A.

It has not been considered worth while to examine the data obtained from the sexual polyploids of the Taynuilt A gradient since only two (1945) and three (1946) populations were available. The polyploids at sea level, however, present a complex picture which merits closer attention. Although the sexual polyploids which were growing on Ben Cruachan were all found to have the same chromosome number, namely (2n = 42), varying counts were obtained from those growing in the quite limited area at sea-level.

TABLE XII.

Population Number	Chromosome Number (2n =)	
	Wild material	Garden Material
8	42	42, 49
9	49, 56	42, 56
10a	42, 49	-
29	42	42
30a	42	42

It is apparent from Table XII that two distinct chromosome races exist and that they are capable of crossing and occasionally at any rate, producing viable hybrids. The problem of their distribution is less easily solved until the biotic environment is examined more closely.

Population 9 where the only octoploids were found, grows actually on and at the base of, a stone wall cemented with mortar

FESTUCA RUBRA. POPULATION 30 : ILLUSTRATION OF DIFFERENCE IN HABIT OF GROWTH BETWEEN (TOP) FIRST-YEAR PLANT
(BOTTOM) SECOND-YEAR PLANT.

and, incidentally, giving a neutral soil reaction. The habitat is dry and well-sheltered by trees and there is an almost complete absence of competition. Population 10a is growing on the side of a dry, grassy bank under the rather intermittent remains of an overgrown hawthorn hedge some hundred yards from Loch Etive shore. The habitat is fairly sheltered save from the North-east which is not the direction of the prevailing wind. Population 8 is growing on the edge of a pasture field just above the River Awe. The habitat is again fairly sheltered with some gorse bushes and with several hawthorn trees very close but the F. rubra plants which were collected were growing in open spaces between the taller vegetation with less overhead shading than those of Populations 9 and 10a. Population 29 grows on the entirely open shore of Loch Etive whilst Population 30a is also growing in an unshaded habitat on a roadside but in strong competition with Arrhenatherum elatius M. & K. Population 29 on the other hand is almost entirely free from plant competition but occupies what is undoubtedly the most exposed habitat. Taking into consideration the phytosocial environment, it would seem reasonable to conclude that the octoploid F. rubra is a race of shaded places with a considerable amount of shelter whilst the more common hexaploid is most suited to exposed areas which would account for its uniformity of occurrence on the hillside. From present evidence crosses between the two races apparently do occur and it seems likley that they are able to grow in habitats intermediate between the shaded and the very exposed. In the present case an examination of the character mean values and the

TABLE XIII.

1945 Polyploid Sexual, F. rubra.

Mean Values and Divergence Indices. Taynuilt, Sea-level.

Leaf Length

DI.

MV.

	9	8	29	30a
9		63	69	71
8	421 438		75	65
29	421 411	438 411		62
30a	421 445	438 445	411 445	

Leaf Breadth

DI.

MV.

	9	8	29	30a
9		67	65	71
8	2.6417 2.8208		67	75
29	2.6417 2.2838	2.8208 ** 2.2838		60
30a	2.6417 2.2113	2.8208 ** 2.2113	2.2838 2.2113	

MV.

	9
9	
8	8.2 8.1
29	8.2 8.0
30a	8.2 7.3

Potential Fertility

DI.

MV.

	9	8	29	30a
9		54	63	54
8	462 200		61	50
29	462 336	200 336		61
30a	462 * 92	200 92	336 92	

Actual Fertility

DI.

MV.

	9	8	29	30a
9		50	54	50
8	153 107		54	50
29	153 199	107 179		54
30a	153 36	107 36	179 * 36	

MV.

	9
9	
8	35.4 23.4
29	35.4 ** 18.1
30a	35.4 ** 14.9

Spikelet Size

DI.

MV.

	9	8	29	30a
9		56	58	60
8	9.0 9.2		56	68
29	9.0 9.0	9.2 9.0		52
30a	9.0 8.5	9.2 8.5	9.0 8.5	

Flowering Grade

DI.

MV.

	9	8	29	30a
9		52	68	50
8	2.9 3.3		66	62
29	2.9 3.3	3.3 3.3		68
30a	2.9 2.2	3.3 2.2	3.3 2.2	

MV.

	9
9	
8	41.7 35.0
29	41.7 35.4
30a	41.7 37.1

1948 Polyploid Sexual, F. rubra
Character Mean Values and Divergence Indices. Taynuilt Sea-level.

Leaf Length
DI.

MV.	9	10a	8	29	30a
9		67	60	60	63
10a	402 * 450		60	58	64
8	402 407	450 * 407		56	66
29	402 421	450 421	407 421		62
30a	402 * 456	450 456	407 * 456	421 456	

Leaf Breadth
DI.

MV.	9	10a	8	29	30a
9		54	54	64	63
10a	2.6000 2.4658		54	58	59
8	2.6000 2.6096	2.4658 2.6096		60	61
29	2.6000 * * 2.1217	2.4658 * 2.1217	2.6096 * * 2.1217		57
30a	2.6000 2.3408	2.4658 2.3408	2.6096 2.3408	2.1217 2.3408	

Plant Weight (oz.)
DI.

MV.	9	10a	8	29	30a
9		75	64	67	72
10a	109 ** 55		71	67	53
8	109 106	55 ** 106		67	69
29	109 ** 69	55 69	106 ** 69		63
30a	109 ** 53	55 53	106 ** 53	69 53	

* SIGNIFICANT AT ·05 POINT.

** SIGNIFICANT AT ·01 POINT.

Divergence Indices (Tables XIII and XIV) does not give much indication of the presence of distinct morphological differences between the chromosome races though, of course, it must be allowed that there is a considerable amount of natural variation which must tend to obscure the true pattern. Böcher (1944) found two chromosome races in Veronica officinalis but states that morphological separation is impracticable (p. 12). However, it is clear enough that in the case of each character, the higher Divergence Indices occur between pure populations of hexaploids and those populations in which octoploids and aneuploids have been found. Again, the statistically significant differences which are present, are between the pure populations and the mixed. The greater degree of similarity which might have been expected to occur between the two hexaploid populations does in fact exist for most characters and only once, in the case of Actual Fertility is there a significant difference between the two. The data for Population 10a (1946) is perhaps a little surprising in its fairly close similarity to the data for Population 30a until it is remembered that the actual proportion of the different polyploid races in each mixed group examined is unknown since only representative plants from each habitat were used.for a chromosome count. A greater number of hexaploid plants among those measured for Population 10a would naturally render the mean value more akin to that for a purely hexaploid population than to that for a more mixed population. For this reason, it is rather futile to attempt to draw final conclusions concerning either the morphological distinctness of

FESTUCA RUBRA. EXAMPLES OF TYPES OF HABIT OF GROWTH.

the chromosome races or the distribution of variation within them. It would be necessary for these purposee to deal only with plants of known chromosome number throughout.

(2) Taynuilt B.

Since only two (1945) and one (1946) populations were available, no reliable conclusions could be drawn from the data though again in a very limited way, altitudinal trends similar to those indicated for F. ovina could be discerned.

(3) Dalmally.

As has already been noted the only chromosome race found among the F. rubra plants on Ben Cruachan has a diploid number 2n = 42, and the plants from the Dalmally gradient are no exception. It seems likely that the populations are individual but that there are no real barriers to continuous gene flow along the gradient since there are none to few significant differences between population mean values for the different attributes. No attempt has been made to calculate Divergence Indices under the circumstances. The altitudinal slope is so gradual that it would not have been surprising to find a significant correlation between character change and altitude but the presence of bracken must be sufficient to erect at least a partial barrier. The reason for the rather erratic variation then becomes clearer for the bracken does not occur regularly but in bands and patches. Again as with the F. ovina populations of the Taynuilt A gradient, these F. rubra populations occur in the area which is only intermittently affected by, for example, snow, and to a much more continuous degree, mist.

TABLE XV.

1945 Viviperous, F. vivipara. Taynuilt B.

Population	Altitude	Leaf Length		Leaf Breadth		Plant Height		Plant Weight		Habit of Growth		Flowering Grade	
		MV	DI	MV	DI	MV	DI	MV	DI	MV	DI	MV	DI
	feet	mm		mm		100s of mm		lb.		ratio			
27	1450	334		.6960		6.7		11.6		1.01		4.8	
		*		**		*				**			
25	2000	278	80	.7729	71	6.1	70	15.1	67	1.56	77	6.5	72
		*				**		**					
23	2400	220	66	.7921	71	5.0	77	9.3	71	1.30	63	7.6	55
						**							
21	2800	251	61	.7371	65	6.3	93	10.3	64	1.03	64	6.8	54
		*		*		**		*		**			
16	3639	178	81	.9746	88	4.3	96	4.6	85	2.30	100	7.6	53
Divergence Index between end points			95		100		97		85		100		76
Significance between end points		xx		xx		xx		xx		xx		xx	
Correlation Coefficient Character: altitude		-.652 xx		+.646 xx		-.021		-.406 xx		+.502 xx		+.468 xx	

* SIGNIFICANT AT ·05 POINT.

** SIGNIFICANT AT ·01 POINT.

TABLE XVI.

1945 Viviparous, F. vivipara. Dalmally.

Population	Altitude feet	Leaf Length MV mm	Leaf Length DI	Leaf Breadth MV mm	Leaf Breadth DI	Plant Height MV 100s of mm	Plant Height DI	Plant Weight MV lb.	Plant Weight DI	Habit of Growth MV ratio	Habit of Growth DI	Flowering Grade MV	Flowering Grade DI
63	400	284		.7409		6.7		12.4		1.09		5.2	
		*										**	
38	1100	337	70	.7333	57	6.4	62	11.8	68	.95	61	8.1	85
												**	
40	1500	313	56	.7575	58	6.4	52	14.4	71	1.26	75	4.5	92
		**				**							
42	1900	415	91	.7031	76	7.6	91	15.6	64	1.09	56	3.6	50
		**				**		*				**	
44	2300	316	90	.8567	80	6.1	84	11.2	68	1.44	69	6.5	71
46	2700	323	56	.8296	64	6.2	54	9.0	58	1.47	56	6.1	50
		**				**							
62	3100	245	79	.9214	85	4.6	86	10.3	57	1.58	60	6.8	59
Divergence Index between end points			68		91		98		69		76		69
Significance between end points				xx		xx				xx			
Correlation Coefficient Character: altitude		-.066		+.602 xx		-.157 xx		-.211 xx		+.611 xx		+.039	

* SIGNIFICANT AT ·05 POINT.

** SIGNIFICANT AT ·01 POINT.

(c) Viviparous polyploid. *F. vivipara.*

(1) On the Taynuilt A gradient no plants were available among the 1945 plants save two populations at sea-level, an insufficient number for any detailed analysis. With the 1946 plants, however, all the populations on the gradient were represented in the trials and Table XVII shows how generally similar is the distribution of variation to that already noted for the sexual diploids on the same gradient. The apparently random distribution of the character mean values is very marked.

(2) and (3) The similarity in the distribution of variation is even more marked on the Taynuilt B gradient and the same is equally true of the populations on the Dalmally gradient.

This strong confirmation of the conclusions which may be drawn from the sexual diploid populations is very satisfactory but it also presents a new and puzzling problem. Though the factors which govern the extent and direction of differentiation may be imperfectly understood, it is nevertheless possible to appreciate that in cross-fertilising species, the distribution of variation is achieved through sexual reproduction. It is much more difficult to understand how differentiation between populations, and between individual plants, comes into being when the plants in question have a viviparous form of reproduction. If the plants are entirely viviparous, there can be no question of any form of gene exchange between individuals and the only possible source of further differentiation is mutation. If, however, the plants even occasionally produce some normal sexual flowers then the possibility of a limited amount of interbreeding

EXAMPLES OF UNUSUAL FESCUE PLANTS.

(TOP) VIVIPAROUS PLANT WITH SEVERAL FULLY-SEXUAL PANICLES.

(BOTTOM) SEXUAL PLANT WITH SOME ENTIRELY-VIVIPAROUS PANICLES.

TABLE XVII.

1946 Viviparous, F. *vivipara*. Taynuilt, A.

Pop.	Alt. feet	Mean Character Value. LL mm	LB mm	H/G Ratio	PW oz.
30	0	302	.8096	1.0	40.6
1	200	287	.7911	1.5	** 28.0
2	400	** 344	.8275	2.0	** 39.3
3	600	** 281	.8050	1.7	36.6
4	800	* 312	.7700	1.7	40.5
5	1000	332	.7500	2.0	43.2
6	1200	** 284	** .8508	2.0	37.5
7	1400	291	.8042	2.2	39.4
14	1600	294	.8213	1.9	36.3
Significance between end points					
Correlation Coefficient char:alt.		-.083	+.029		

* SIGNIFICANT AT ·05 POINT.

** SIGNIFICANT AT ·01 POINT.

TABLE VIII./

TABLE XVIII.

1946 Viviparous, F. *vivipara*. Taynuilt B.

Pop.	Alt.	Mean Character Value			
		LL	LB	H/G	FW.
	feet	mm	mm	Ratio	oz.
27	1450	334	.7946	2.0	40.7
26	1800	** 262	.8129	2.1	40.3
25	2000	234	.7767	2.4	38.5
24	2200	264	* .8271	2.6	36.2
23	2400	* 295	.8104	2.0	37.7
22	2600	* 261	.8500	2.1	* 31.2
21	2800	247	.8583	2.1	34.4
20	3000	241	.8996	2.3	36.5
19	3200	241	** .8146	2.6	38.4
18	3400	247	.7758	2.8	** 28.4
17	3600	234	** .8875	2.7	31.3
16	3689	234	.8850	2.5	26.8
Significance between end points		xx	xx		xx
Correlation Coefficient char:alt.		-.386 xx	+.214 xx		

* SIGNIFICANT AT ·05 POINT.

** SIGNIFICANT AT ·01 POINT.

TABLE XIX.

1946 Viviparous, _F. vivipara._ Dalmally.

Pop.	Alt.	Mean Character Value			
		LL	LB	H/G	PW
	feet.	mm	mm	Ratio	oz.
63	400	260	.7917	1.5	41.0
35	500	* 307	.7633	1.0	45.2
36	700	** 232	.7783	1.0	** 33.6
37	900	261	.7967	1.0	34.9
38	1100	259	.7721	1.1	* 43.1
39	1300	244	.7671	1.0	** 34.2
40	1500	222	.7463	1.0	35.0
41	1700	** 340	** .8225	1.5	40.4
42	1900	** 295	.8167	1.7	42.1
43	2100	** 251	.8408	1.7	40.3
Significance between end points			xx		
Correlation Coefficient char: alt.					

* SIGNIFICANT AT .05 POINT.

** SIGNIFICANT AT .01 POINT.

with consequent gene recombinations provides an important source of differentiation. Turesson found in his investigation of the viviparous fescues that only two northerly districts had an entirely viviparous form while the majority of the largely viviparous forms, which included specimens from Scotland, produced a few weak shoots late in the season bearing some apparently normal flowers. Some had all the floral parts but others had only some parts and some were entirely rudimentary. Jenkin (1921) also found that similar hunger inflorescences were produced late in the season by viviparous plants from Wales and viable seed was actually obtained under garden conditions. The Ben Cruachan fescues behaved in a similar manner but though apparently abundant pollen was formed and some flowers did appear to be entirely normal, no seed was found under garden conditions. None of these late shoots were ever observed under wild conditions but the possibility of seed being produced cannot be ignored.

There does not seem to be much possibility of interbreeding between _F. ovina_ and _F. vivipara_ since the inflorescences on the latter were not produced, under garden conditions at any rate, until the _F. ovina_ plants were well past the flowering stage. It must be admitted that another serious objection to the production of mature seed by these hunger inflorescences arises from the fact that they are produced so late in the season that they must have a very brief period favourable for seed ripening. Since differentiation is so marked in the viviparous fescues, however, it seems necessary to postulate such a source of gene recombination in addition to the presence of mutation

FESTUCA VIVIPARA. EXAMPLES OF TYPES OF PANICLES FROM TWO SINGLE PLANTS.

(TOP) WHOLLY VIVIPAROUS AND WHOLLY SEXUAL.

(BOTTOM) WHOLLY VIVIPAROUS AND PARTIALLY SEXUAL.

and it may be that the limited and intermittent amount of crossing which is possible, accounts for the more marked character of the variation. If variants are only produced at irregular intervals, the pressure of natural selection may be allowed an unusual length of time for eliminating individuals which show unsuitability for the environment whether to a greater or a lesser extent.

Heribert Nilsson (1947, p. 141) has advanced the theory that polymorphy in a truly apomict species is a sign of senescence. He argues that when the degree of apomixis becomes absolute, there are probably millions of biotypes within such a species but the pressure of natural selection gradually reduces this number, finally to a few hundreds and it is when the number becomes so small that their existence becomes most obvious to the taxonomist. It has been postulated that the mutation rate decreases with the increasing age of a species and what mutations occur, would not influence the few definite biotypes to any appreciable extent.so that they would be very distinct from each other. This seems to be a reasonable theory and would fit the present case if it were not for the observed ability to produce some sexual flowers even though their presence under natural conditions has not been noted.

There is a further point which makes it impossible to regard the Ben Cruachan F. ovina vivipara plants as completely viviparous, namely the production of apparently normal, entirely sexual plants from viviparous bulbils. A completely viviparous plant 13E was collected on Ben Cruachan and kept in the greenhouse for cytological investigation. It was not remarkable in any way and a root-tip count yielded the normal diploid number of $2n = 28$.

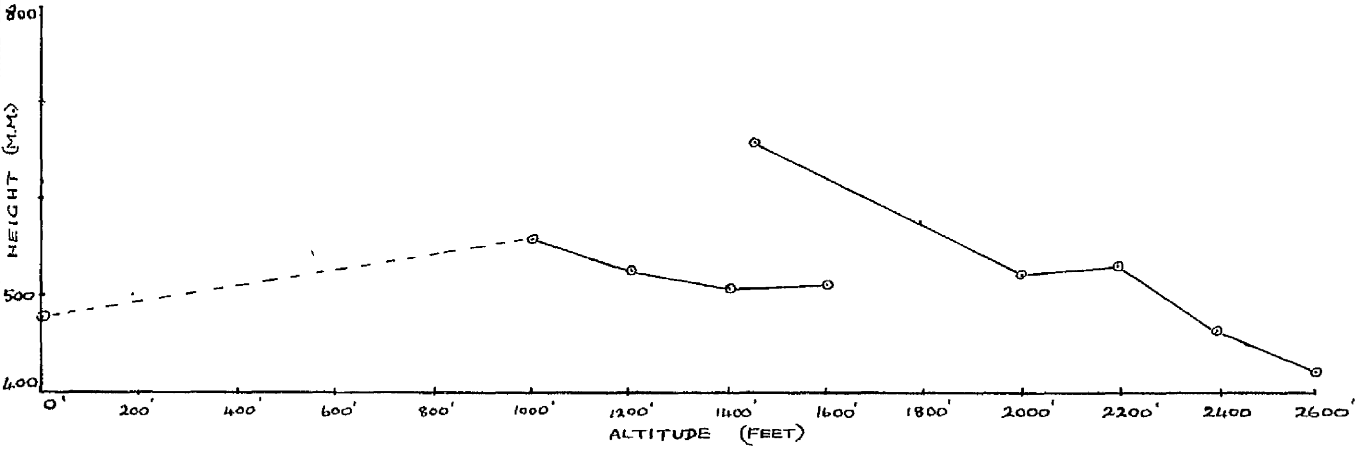
60a

FESTUCA VIVIPARA. EXAMPLES OF TYPES OF HABIT OF GROWTH.

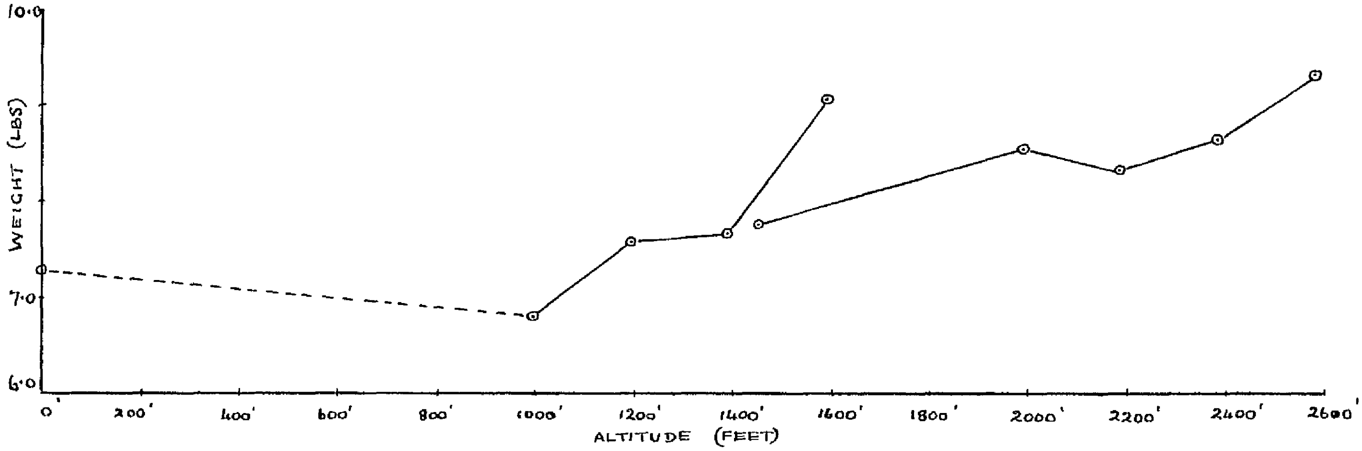
In the second season, however, a semi-viviparous panicle was produced and some apparently normal seeds were formed. The bulbils from the inflorescence were planted in one pot, the seeds in another and though the latter failed to germinate, the bulbils rooted and eventually became normal sexual plants which have remained sexual each season. Jenkin (1921) reported that viviparous plants grown from bulbils produced some partially sexual inflorescences in the second season. Seed obtained from them germinated and yielded four plants, two of which were sexual and two viviparous. Instances have been given of vivipary modificatorily induced but in each of the above cases, healthy, normally sexual plants developed from viviparous bulbils. It would seem that F. vivipara as found in Great Britain at any rate, is not completely viviparous and that some further investigation is required to elucidate the problems of the relationship between the sexual and the viviparous, the question of the change in chromosome number being not the least.

A point of interest arises in connection with Plant Weight and Plant Height measurements for the sexual diploids. The graph for Plant Height 1945 (Graph IV(a)) shows a decrease in mean value as altitude increases but Plant Weight (Graph IV(b)) shows an equal tendency to increase and this tendency is particularly marked for the Taynuilt B gradient. Plant Weight for 1946 plants (Graph IV(c)), however, decreases (with one exception) as altitude increases. The discrepancy between the Weight figures for the 1945 and 1946 plants is probably to be accounted for by the fact that in the first case, the total weight of the plant was recorded

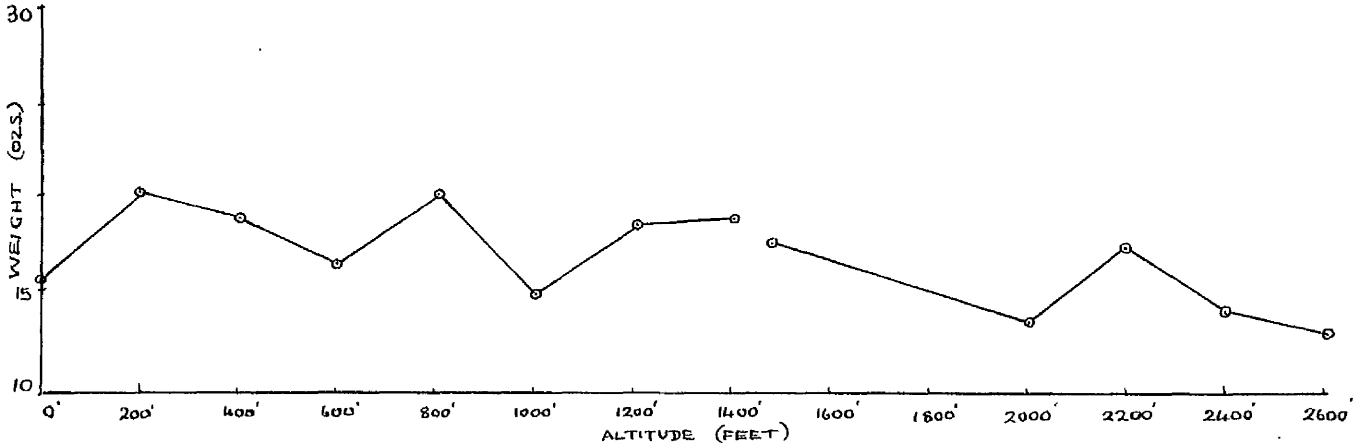
GRAPH IV. TAYNUILT GRADIENTS A AND B



(a) HEIGHT OF DIPLOID SEXUAL (1945) PLANTS.



(b) TOTAL WEIGHT OF DIPLOID SEXUAL (1945) PLANTS.



(c) WEIGHT (SHOOT PORTION) OF DIPLOID SEXUAL (1946) PLANTS.

whilst in the second case, only the shoot portion was weighed. It must be, therefore, that at higher altitudes the root system of the plant increases at the expense of the shoot portion. Salisbury (1940) has noted that "depth of rooting which has been shown to vary between strain and strain may determine drought-resistance or survival from potential 'frost-lifting'." Such attributes would be of great value to plants at high altitudes.

The validity of such an interpretation is rendered dubious, however, by the fact that with both 1945 and 1946 viviparous plants Plant Weight decreases with increasing altitude in spite of the different methods of weighing adopted in the two years. It may, however, point to a general physiological superiority of the tetraploid viviparous type over the sexual diploid at high altitudes.

The data which have been presented in the foregoing tables are sufficient to show that the variation which has been examined is of a hereditary nature. It is true that, for example, the prostrate habit of growth of populations from the higher altitudes had been exaggerated in the wild by the severe environmental conditions but it is obvious that an inherent prostrateness is, nevertheless, included within the constitution of the individuals from these levels. The Analysis of Variance calculations have made it clear that with very few exceptions and those mainly among <u>F. rubra</u>, the Ben Cruachan fescue populations cannot be regarded as a single homogeneous unit. Statistical significance is thus added to the conclusions which may be drawn from eye observations and detailed measurements. Again, the results from two separate seed collections from the natural populations but grown in successive years, are fairly similar. Complete agreement would have been more open to question than are the dissimilarities which have appeared since in each year the seed was collected at random from the populations and must necessarily represent somewhat different samples of the internal variation in each case.

VIII. DISCUSSION.

It has already been mentioned that the aim of experimental taxonomy is to elucidate further the relationships within the most valuable classifications of the systematists, to add to, rather than to supplant them. In any case, it would certainly not be possible to attempt to fit the systematists' units neatly into those proposed as a result of experimental investigations upon some species without many more experiments upon many more species. It only leads to confusion when writers who have not been able to carry out adequate experiments or, perhaps, any at all, hastily apply terms which presuppose a thorough knowledge of the genetical constitution and relationships of the organisms. These fundamental requirements are well stressed by Turrill (1946). Hutchinson, Silow and Stephens (1947) have been able to adopt a classification of Gossypium into taxonomic sections, species and varieties which they say, closely resemble the coenospecies, ecospecies and ecotypes defined by Clausen, Keck and Hiesey (1939) but this has only been made possible by detailed genetical examination in the course of which several groups previously thought worthy of specific rank, were merged into one.

The validity of the morphological criteria which are used to define species require close analysis for the morphological differences which had been used to distinguish these "species" were found by experimental methods to be "genetically simple and unimportant". On the other hand, in the case of Mountain Timothy, the diploid closely resembles the tetraploid in morphological details yet on the grounds of the cytological, genetical and ecological differences, Nordenskiöld (1945) has considered that they ought to be given separate species rank and in the light of experiment Gregor (1946) agrees. Morphological criteria, therefore, may in themselves be unreliable guides to the true relationships between organisms but they have persisted as the main means of discriminating between species and those groups which were accorded specific rank by Linnaeus, have remained largely unaltered to the present day. It is worth noting in passing that even though biologists have failed to agree upon valid, universally-accepted criteria for the species concept, it has always been felt to be a more definite natural group than any other to which taxonomic rank has been given, hence, probably, the general acceptance of the Linnean groupings. Nevertheless, the concept of the species and the idea of its relationship to other groups has changed fundamentally from the original. In the time of Linnaeus species were thought of as specially created entities, final and immutable and only requiring careful enquiry to become clearly defined to the observer. This static concept, however, has given place to the idea of the species as a dynamic level, a stage in evolution which can not only be defined in the present state but, through experiment, may even be traced in a certain degree to its origins.

Nevertheless, the exact method by which the species level has been reached is not a matter of universal agreement. Du Rietz (1930) defined the species as "the smallest natural populations permanently separated from each other by a distinct discontinuity in the series of biotypes". Such an adequate yet neutral definition can be accepted by all but whereas Goldschmidt (1940) holds that species are formed by large systemic mutations, the majority of biologists are convinced that speciation is wholly the result of micro-evolution, the slow accumulation of gene mutations and chromosomal reconstructions under conditions of varying environmental pressure leading to changes within a potentially continuous population. Goldschmidt does not question this micro-evolution below the specific level but believes that, at and above this level, an essentially different process, macro-evolution, takes place. It has not been proved so far that micro-evolution differs in kind from macro-evolution but it seems reasonable to suppose from the evidence so far available that the difference is only one of degree. Hutchinson, Silow and Stephens (1947) consider the possible path of evolution of the convoluted lint hairs and the annual habit in Gossypium and come to the conclusion that since many morphological characters are involved in each, nothing less than an "extensive rebuilding of the genotype" would be able to cause such related characters to evolve. "It is evident", they continue, "that this reorganisation was accomplished by the accumulation of gene differences and not by any 'macro-evolutionary' change of the type postulated by Goldschmidt (1940)". Whatever the mode of evolution of these important characters in *Gossypium*, and the arguments of these authors merit acceptance, even if the production of a new species, genus, or higher category by large systemic mutations could be proved, it would, as Simpson (1944) says, supplement, not supplant, the population theories that involve selection acting on unit deviations of variable but usually small degree.

Timofeeff-Ressovsky (1940, p.127) says "although it is very difficult to give a definition of the species which would be of general value and of practical use in all groups of organisms, we nevertheless know that a high degree of biological isolation is the most important criterion." This biological isolation which is such a vital factor in promoting discontinuity, may take several forms of which the most obvious are those which prevent interbreeding between species colonising the same area. Anderson and Sax (1936) consider that it is seasonal isolation which prevents interbreeding between *Tradescantia hirsuticaulis* and *T. subasper* var. *montana*, two species which may inhabit the same areas but rarely produce hybrids. The fundamental importance of genetical barriers between species is emphasised by Hutchinson, Silow and Stephens (1947, p.12) who illustrate their point with the example of *Gossypium herbaceum* var. *acerifolium* and *G. arboreum* which are deliberately grown in a mixture in certain parts of India. Vigorous and fully fertile

F_1 hybrids are formed as a result of free intercrossing but later generations are feeble, obviously genetically unbalanced and finally fail to survive. The short-term, economic advantage of this policy is obvious but so also is the existence of a genetical barrier which does not prohibit crossing but does prevent the loss of identity of the two species.

There are, also, many instances in botanical literature of species which are capable of modified interbreeding where opportunity permits, though each retains its distinct specific characters save only in a narrow zone at the immediate point of contact where the hybrids are formed. An example is given by Cockayne (1923) in the case of Myrtus bullata and M. obcordata in New Zealand; only M. obcordata is found in South Island up to lat. 41° and in North Island north of lat. 35° M. bullata occurs alone save for one small isolated area where M. obcordata occurs. Between lat. 41° and lat. 35°, however, both species occur and there is a highly polymorphic hybrid population between M. bullata and M. obcordata. It would almost seem that these two species ought to be called subspecies in the sense of Du Rietz (1930) who defines a subspecies as "a population of several biotypes forming a more or less distinct regional facies of a species" were it not that he envisages a continuous intergradation between subspecies whereas in the above case, there is a sharp delimitation.

Huxley (1939) has spoken of the species as the "lowest group-category between members of which fertile interbreeding either is impossible or normally does not occur" but, apparently, would accept M. bullata and M. chocrdata as separate species in spite of the recognisable, recurrent interbreeding, since he says (1938) "the progress of taxonomic research has brought to light numerous cases ------ of the incomplete discontinuity of species". Possibly an explanation of this difficulty of making a universally applicable definition may lie in the degree of stability which has been reached within the gene complex of the species. Experimental work by Timofeeff-Ressovsky and theoretical investigations by Fisher and Sewall Wright have emphasised the importance of the process of building-up of gene complexes, harmoniously stabilised in different ways, within populations which have become isolated from each other. If, in the further process of expansion or migration, two such populations come to colonise the same area but are unable to form viable hybrids because of the evolution in each of distinct, finely-balanced genetic constitutions, such populations are obviously full species. They would also qualify for the rank of coenospecies according to the definition given by Gregor et alia (1936) who define it as "separated from all other plants by sterility or by failure of hybrids to produce viable seed". F. rubra and F. ovina are examples of such species since, as has already been noted, they are found in close proximity without interbreeding. This is not entirely due to seasonal isolation although the time of flowering of F. rubra is at least one week later than that of F. ovina and this may have played an important part in earlier stages of differentiation, but even where crossing has been attempted artificially (Jenkin, 1934) only a small percentage of viable seed was formed and no plants were obtained from them. Such an example is in perfect agreement with Dobzhansky's definition (1941, p. 373) of the species as "that stage of evolutionary process at which the once actually or potentially interbreeding array of forms becomes segregated in two or more separate arrays which are physiologically incapable of breeding". The case of Myrtus bullata and M. obcordata does not agree with this definition, however, and it must be concluded that the stabilisation of the gene-complex of each species has not proceeded so far as to inhibit all interbreeding, yet far enough to permit the groups to acquire specific qualities of distinctness and to retain them under conditions where interbreeding is possible. In such cases where a high degree of equilibrium has been attained, the hybrids will be in a much less advantageous position owing to the upsetting of the harmony achieved by the parent species. Recurrent interbreeding is possible but the hybrids will not be able to compete with the parents and even if they survive, a situation such as Hutchinson, Silow and Stephens have observed in Gossypium herbaceum var. acerifolium and G. arboreum is likely to result.

An excellent example of the evolutionary importance of balanced genomes may be drawn from recent work by Valentine (1947) on hybridisation between the Primrose, Primula vulgaris Huds. and the Oxlip, P. elatior Schreb., the chromosome number in both cases being (2n = 22). With the Primrose as female parent about 85 per cent of the seeds, which are unusually small, contain embryo and some endosperm but the germination is very poor. On the other hand, with the Oxlip as female parent, only some 20 per cent of the seeds produced contain embryo and endosperm but show fairly good germination. The author believes that the difficulty in obtaining viable hybrid seed is largely due to a lack of balance between the genomes of the Oxlip and of the Primrose, which causes an early degeneration of the endosperm. Where the hybrid seed germinates, the F_1 plants are normally vigorous, produce viable pollen and mature germinable seeds, a fact which the author concludes is due to the possession, through first-generation selection, of genetically well-balanced endosperm. Further work on the situation under field conditions has been promised, but since the Oxlip is confined to a small area in East Anglia with Oxlip-Primrose hybrids occurring on the perifery, it appears certain that the lack of genetic balance in the majority of the hybrids provides an effective barrier to the swamping of one or both species.

A concept of the species which would be adequate to define species of the type of Myrtus bullata and M. obcordata in their relationship to each other at any rate, would be almost akin to that of the ecospecies which is defined by Gregor et alia (1936) as "separated from other parts of its coenospecies by restricted interfertility or by failure of hybrids to establish themselves in Nature". Hybrids are able to establish themselves at least to a limited extent, however, and in some cases to persist for a few generations so this is an instance of the difficulties which arise when an attempt is made to equate taxonomic categories with the units of experimental taxonomy.

To summarise briefly, the species of the alpha taxonomist would seem to include groups showing degrees of discontinuity varying from complete to fairly high, and where the line is drawn must still depend largely upon morphological details in the absence of experimental data. Apparently the taxonomic species may include both the coenospecies and the ecospecies according to their definitions from the genetical point of view.

A lesser degree of balance may be envisaged within the gene-complex of the subspecies as defined by Du Rietz (1930). The chief isolating factors in the case of this taxonomic unit are geographical or it may be ecological, and where such barriers are removed, interbreeding is fully possible. Under natural conditions, however, intergradation between subspecies is frequently found to occur. Du Rietz (1930) gives an example of the continuity between subspecies in the case of Celmisia petiolata Hook. in New Zealand where he distinguished at least three subspecies each with a distinct regional distribution in the Southern Alps but continuously intergrading the one with the other. Where intergradation is not immediately obvious, however, subspecies may be thought to be worthy of species rank. Cain (1944) quotes the instance of the titmouse Parus major which extends all over Europe, south to Iran and across Siberia to Amur; P. bokhariensis extends over southern Asia and intergrades with P. major in Iran and with P. minor in South China; P. minor extends from South China to Amur where it occurs with P. major without interbreeding. In this example of a "ring of races",

P. major and P. minor could be ranked as separate species if it were not known that intergradation exists between them through P. bokhariensis. Further investigation in the field may bring to light many more instances of this type.

The emphasis is obviously upon the importance of geographical isolation. No doubt in the last example, P. major and P. minor have come to share the same area in Amur in comparatively recent times after separation sufficiently prolonged to make interbreeding impossible. Similar isolation may in time break the chain of intergradation between the two subspecies through P. bokharienses and as small gene changes gradually accumulate, an equilibrium may be established within the respective genetical constitutions rendering crossing ineffective.

Dobzhansky (1941), however, considered such an explanation of the formation of an isolating barrier by the accumulation of gene changes to be genetically unsatisfactory and tentatively suggested that "once an incipient physiological isolation has become initiated, natural selection will tend to strengthen it and eventually to make the isolation complete". That this may indeed be the case has been shown by Stephens (1946) in the case of recently-differentiated species among the New World cottons, Gossypium hirsutum var. marie-galante and G. barbadense L. The interspecific barrier is rendered effective by means of an isolating mechanism which depends on alleles occurring at one locus only so that no seeds develop as a result of interbreeding. Although only some types are "carriers", Stephens says that those carrier types "in a mixed barbadense and marie-galante population will have a selective advantage and will ultimately replace non-carriers."

The importance of genetical isolation has already been made plain but it is difficult to see how such isolation could be achieved under sympatric conditions and it would seem almost essential to postulate some form of initial spatial separation unless a major systemic mutation such as that envisaged by Goldschmidt were allowed or, perhaps, a change in ploidy. It is natural to think primarily of geographical isolation and its importance is undisputed. An illustration may be taken from the clear analysis by Crocker and Wood (1947) of the initiation of isolation through the Recent aridity in South Australia and its effect upon plant communities. Species were forced by the sudden climatic change into ecological refuges whence they gradually recolonised denuded areas whenever more suitable conditions prevailed, but large areas remained without vegetation. Crocker and Wood are of the opinion that a considerable amount of speciation has already taken place in widely-separated habitats but that, nevertheless, the period of time which has elapsed since the beginning of the arid period has been too short for much progress to have been made in this direction. The speed with which speciation may occur is dependent upon a number of factors and under the conditions prevailing in South Australia, two of the most important must have been the edapho-climatic range of tolerance of a species, its powers of migration and the frequency of the occurrence of mutation. Species such as Eucalyptus oleosa and E. gracilis with a wide range of tolerance were able to form relatively wide and continuous distributions through successful migration from several refuges and little differentiation appears to have occurred. On the other hand, *E. cladocalyx* is found in three separate localities in South Australia, each the result of a slow migration from a single survival habitat but in spite of this continuing isolation, no major variation is reported between the three localities although there are differences in vigour probably due to edaphic conditions. In the case of *Acacia Sowdenii* Maiden and *A. Loderi* Maiden, however, taxonomically separate species have been recognised. They occur in areas at a considerable distance from each other and the authors are of the opinion that they have been derived from a common ancestor but are inclined to consider that the differentiation has scarcely proceeded far enough to warrant separate species rank.

A further example of a somewhat similar nature is provided by Anderson (1936). This author has put forward the interesting and well-supported theory that *Iris versicolor* is a pre-glacial or inter-glacial hybrid between *I. virginica* and *I. setosa* var. *interior*. At the end of the glacial period the hybrid was left in a more youthful area which Fernald (1931) characterises as "the vast region of Canada and the Northern States which has become available for wholesale occupation by plants only since the decay of the Wisconsin ice, within the last few thousand years." Here, through geographical isolation from the parent species, the hybrid was able to build up a stabilised and independent gene-complex so that nowadays it can colonise the same area as *I. virginica* with only a limited amount of interbreeding.

It must be remembered that geographical isolation does not necessarily mean wide spatial separation and Mayr (1947) says "the term 'geographically isolated' --- refers to any environmental factor that effectively inhibits gene flow between two neighbouring populations". The isolation created by preference for different ecological habitats is given prominence in this statement and such isolation would certainly seem to exist in the example given by Du Rietz of the different subspecies of *Celmisia petiolata* in New Zealand. Turrill (1938) has examined in detail the case of *Silene alpina* and *S. Cucubalus* which grow in adjacent habitats in the French Alps with a minimum of interbreeding. Even more striking examples may be found in recent American work on fishes. Lagler and Bailey (1947) found that two races of the Johnny darter, *Boleosoma nigrum*, have distinct geographical distributions but also occur within the same area whilst still retaining their characteristics through their preference for different ecological habitats. *B. nigrum eulepis* Hubbs and Greene is found in quiet, slow-moving waters with a rather loose mud or silt bottom and a fair amount of aquatic vegetation; *B. nigrum nigrum* on the other hand, prefers water where there may be moderate wave action and where the bottom is somewhat firmer. The two subspecies occur together in some streams in Northern Ohio but are segregated in the breeding season when *B. nigrum nigrum* prefers to move upstream. The subspecies retain their attributes under the uniform environmental conditions of captivity and are capable of free interbreeding. Intergrading populations have been found and show that interbreeding is equally possible under natural conditions were it not that habitat preferences keep the subspecies apart. Lager and

Bailey quote a further example from Trautman (1931) of Notropus volucellus. Three subspecies are found in three habitats, small streams and lakes, quiet backwaters or large rivers, open channels of the rivers, within a single area and though there is intergradation at the boundaries of the habitats, the subspecies still remain discrete.

The ecological isolation of subspecies is not always as apparent as it is in the above examples, or in the case of species, because of the continuous though diminished gene flow but Mayr (1947) says "In much of the current literature, the subspecies and the ecological race, are treated as two completely distinct phenomena. The truth is, however, that they are merely two facets of a single phenomenon." This is very true when it is considered that every population is basically an ecological one otherwise it could not tolerate the given conditions and would not exist.

Further examination of the lower units leads to the conclusion that since the fundamental basis of the ecospecific concept is restricted ability to exchange genes and there need not be an explicit ecological or regional criterion, the subspecies in Du Rietz's sense cannot always be equated with the ecospecies. Indeed, the continuous intergradation which so frequently exists among subspecies makes this unit bear a closer resemblance to the ecotype which is not separated from other ecotypes by the barrier of sterility. It is possible, therefore, for a subspecies to be called quite correctly an ecotype and this has been done by Clausen, Keck and Hiesey (1940) in the case of

Potentilla glandulosa. Since "ecotypes are not always morphologically distinct" (ibid p. 32) however, and subspecies as systemic units are, the two terms are not necessarily interchangeable.

Some mention may be made at this point of an attempt to classify agricultural crops on a geographical-ecological-morphological basis which is to be found notably in the work of some Russian authors and which introduces completely new terms such as proles and grex. The limitations of knowledge of the language makes difficult an accurate understanding of the concepts involved but a general inference may be drawn at second-hand from the work of Schiemann (1946). It would appear that the proles is conceived to be an ecological-morphological-regional population below the taxonomic rank of subspecies and may contain greges which are geographically distinct groups (also spoken of as groups of varieties) with distinct morphological characters. Subproles are also mentioned although it might have been thought that greges would be sufficient. The names given to various proles, to those belonging to Triticum aestivum ssp. indo-europaeum by Flaksberger (1935), for example, suggest a curious mixture of geographical-ecological criteria whilst on the other hand, the names given by the same author to the proles of T. aestivum ssp. irano-asiaticum suggest a purely morphological basis. The barleys have been similarly classified by Orlov (1936) (quoted by Schiemann) and though the names of the proles suggest morphological criteria, they are consistently classified on a geographical or regional basis.

A much more full understanding of the concepts is required before any criticism can be made but it may be said now that it seems unfortunate that a further series of terms should be introduced to complicate the classification of infra-specific groups unless it is felt that the artificial conditions of crop plant evolution render inadvisable an application of those terms used hitherto for natural populations. Nevertheless, if this is the intention behind this classification ecotype is also employed though its definition is not clear. Ciferri (1944) who mentions these terms freely actually equates ecotype with grex in several instances but in other cases appears to consider it a more general and wider term than proles.

Many systematists recognise in the variety a rank lower than the subspecies but one with a local rather than a regional application. Du Rietz defines it as a "population of one or several biotypes forming a more or less distinct local facies of a species" and also refers to it as a "local race" and "the lowest unit in the series of taxonomical units forming more or less closed inter-crossing populations or syngameous". If varietal rank were always used with such meanings and if a definite ecological basis were present, there is no reason why the term ecotype might not be applied to the systematist's variety. Huxley (1939a), however, would prefer to see 'variety' abandoned altogether as a taxonomic term on account of the many conflicting interpretations given to it. Taken in Du Rietz's sense, it would seem that the variety implies a lesser degree of discontinuity than the subspecies but varietal rank has been conferred on the purely morphological grounds of a few characters and if such a principle were followed to its logical conclusion, the number of varieties would be infinite. Some of the difficulties involved in a purely morphological classification have been emphasised by Skalinska (1947) who notes that in the case of "a great number of characters of the leaves and flowering stem" of Valeriana officinalis L. the morphological analysis of a number of strains from nature has shown that these characters are not regularly correlated, but, on the contrary, they appear in a variety of combinations. In view of this, any attempt to subdivide this polymorphous species into smaller units on a morphological basis must result in a failure. This is just what has been done, however, particularly below the subspecific level in many species, and the result is not classification but greater confusion. Yet it is obvious that such hereditary character differences are of importance, although the degree of discontinuity is small, and could provide a valuable key to the evolution of the species if the correct method of interpretation could be found.

Huxley (1938, 1939b) proposed the concept of the cline and it would appear that this is a possible means of evaluating particularly those small differences occurring at the infraspecific level, differences which it is as fatal to true understanding to ignore as to treat as valid classificatory bases.

The cline is defined very simply as a "gradation in measurable characters"(Huxley 1938) and prefixes may be added to denote the type of cline which is involved under any given conditions. "Ecocline" may be used where the gradient passes through a succession of ecological zones or habitats whilst "genocline" is applicable where a gradient in genes can be detected. The "topocline" has been defined by Gregor (1939) as "a character gradient tracing an essentially geographical course". The cline concept then obviates the disadvantage of naming forms which when examined by themselves appear from the morphological point of view to be distinct but which in actual fact are only more obvious points on a gradient of change within a group. Huxley gives several interesting examples of size clines in relation to birds notably wing length and tarsus length in the wren, Troglodytes troglodytes. By means of the cline, it has been possible to demonstrate a North-South gradual increase in size of these characters from Iceland to Britain passing through the Faroes, Shetlands, and Fair Isle. Different subspecies had already been named for each area save Fair Isle where the differences are so small that no one has felt tempted to accord subspecific rank. The recognition of the cline, however, shows the variation of the Fair Isle population in its true perspective and is of great biological value. By this method, therefore, hereditary variation of whatever type, may be examined in detail and related to similar variation in other areas thus enabling an idea of the pattern of differentiation in response to varying environmental, or other, factors to be obtained. All individual characters do not necessarily follow the same cline or are influenced by the same factors and that an application of the concept to Valeriana officinalis might have fruitful results is shown by a further quotation from Skalinska's paper.

"In populations exhibiting genic differentiation, the various forms may show some habitat preferences. If they occur at the limit of two different habitats, a gradual topographic separation of two biotypes with a different genic composition may take place. Presumably such a process is at work in the mixed population at Pinchley Wood, Glos. where different external factors seem to favour a selection in two opposite ways."

The cline concept has been successfully applied by Böcher (1944) on an ecological basis to leaf size in Veronica officinalis. He found that there was an increase in size on a cline extending from open habitats through intermediate areas into woods. In this case only the single character of leaf size was examined, but in the case of Viola Riviniana Rchb. Valentine (1944) traced an ecocline involving subspecies in respect of general characters. Subspecies minor is common in rather exposed areas such as cliff tops and open heaths whilst subspecies nemorosa prefers woodland habitats and sheltered hedgerows. Between these two subspecies a forma intermedia has been distinguished and this is found to occupy habitats which are intermediate between the sheltered and the fully exposed. Furthermore, a topocline may be found running east and south to the Balkans where subspecies neglecta W. Becker is found. A further example of the application of the cline concept is given in the paper on leaf variation in Asclepias tuberosa by Woodson (1947) who distinguishes a genocline for LA and another following the same direction but not of coincident extent, for LB, between the subspecies, A. tuberosa interior and A. tuberosa tuberosa. Median length and median width provide a "series of rather poorly defined ecoclines," the leaves in the north being in general longer than those in the south. The nomenclature is in doubt here since as Woodson himself says "environmental effect can be discounted apparently because the lines run contrary to the ecocline contours for length" and suggests that the courses of the lines cannot "indicate other than hybrid vigour" while the restriction of population affects the course in the S.W. It might be better to call it a topocline.

It has already been noted that different degrees of discontinuity are the bases of separation into taxonomic groups at and below the species level. Below the subspecific level, the degree of discontinuity diminishes and the continuity is on the whole more striking than the differences though where the emphasis is placed depends largely upon the point of view of the observer. The systematist, accustomed to classifying variation and arranged organisms into groups on points of difference, is inclined to concentrate upon evidence of divergence rather than of continuity between units. Workers in the field of experimental taxonomy are not all agreed on this question and Turesson, perhaps because he collected much of his material from extremely diverse and widely separated habitats, is of the opinion that habitat-groups are sharply delimited. Gregor (1944) however, in an examination of data presented by Turesson (1922) on Hieracium umbellatum finds undoubted support for the more widely-held opinion that changes take place gradually and emphasises that the relative frequencies of the three growth-habit types in the three habitat areas show that a definite intergradation is actually to be found.

If discontinuity were the more striking and universal condition, the concept of the cline would not be so readily applicable yet the foregoing examples of reputed clines and many more which could be quoted, present reasonably valid evidence for the existence of a large measure of continuity in variation.

Further possible clines may be detected in the paper by Turesson (1926) where he observed that in specimens of Festuca ovina L. collected from different parts of Sweden, those from the South had smaller spikelets than those from Central Sweden while the latter were in their turn, succeeded in the North by populations of plants with still larger spikelets. Here, then, is an example of a definite and continuous gradient of change related presumably to climatic conditions but it is a change for which it is difficult to give a reasoned explanation unless it is regarded as the phenotypic expression of linkage between the gene for spikelet size and genes influencing some physiological aspect of the plant. This is a somewhat lame interpretation, however, and it might be more accurate to consider the gradient as a topocline. Turesson also found, however, that there was a gradient of increasing earliness from South to North so it could be that spikelet size is closely linked with earliness in the gene complex but proof would be required. A corresponding variation in earliness was found between two populations from a mountain in Lappland where the population at the foot was later when grown in the experimental garden than that from a higher altitude though the distance between the two habitats in the wild was only 400 metres (1312 ft.). Unfortunately no measurements were given for spikelet size for these two populations so no comparison with the Ben Cruachan dipoiids was possible. Exceptions to the gradual change in earliness do occur but they do not lessen the validity of the gradient since in both instances mentioned, the populations occupy very definite habitats; one the alpine region beyond Northern Sweden, the other, the "alvar" (calcareous rock) of the island of Öland off the S.E. coast of Sweden.

Sinskaja (1942) criticises Turesson for the belief in discontinuity which he maintains in spite of such observations (1926) quoted above, and gives many examples from her own work and that of other Russian biologists in support of the cline which she considers to be a very valuable auxiliary concept. She has added further auxiliary concepts in an endeavour to clarify the somewhat confused position which exists below the lowest taxonomic unit which she considers allowable, namely the subspecies, with which she identifies "large ecotypes differentiated not only according to their biological and physiological characters but morphologically as well." The most important concept is that of the "eco-element" which is "the first stage of divergence which grows wider as the phylogenetical process proceeds" and is further defined as " a morphological and biological constitution formed within the population." The emphasis is upon the entire constitution not upon individual attributes and this is made plain in her theory of spirally overlapping ecoelements a representation of which is given below

$$\begin{array}{cccccccc}
a^1 & a^1 & a^1 & a^2 & & & & \\
 & a^1 & a^1 & a^2 & a^2 & a^2 & & \\
 & & a^1 & a^2 & a^2 & a^3 & a^3 & \\
 & & & a^2 & a^3 & a^3 & a^3 & a^3
\end{array}$$

If a^1 is thought of as an erect ecoelement a^2 as medium and a^3 as prostrate, it will be seen that the figures 1, 2 and 3 represent the hebit of growth which incidentally is being used as an "indicator" attribute, and this in the diagram shows a gradual change from the erect to the prostrate but Sinskaja rightly emphasises that the letter "a" signifies not the individual characters but the ecoelements in their entirety. Thus a^1 may be an erect ecoelement at each level but the "a" constitutions are not, therefore, identical but merely analagous. This may be accepted as correct even in the absence of proof which indeed would be practically impossible to obtain in view of the difficulty of examining any population in its total entirety. Sinskaja does not mention the cline concept in relation to this type of variation but her spiral is closely parallelled by the internal cline.

The "isoreagent" is another auxiliary concept used by Sinskaja to cover chance deviations of no adaptive significance occurring under "unstable environmental conditions". Yet at another point, she says that highly specialised ecotypes do not include ecoelements which can be appreciated, but may have one or two iso-reagents. A contradiction seems to appear here since it does not seem reasonable to expect to find a highly specialised ecotype under such environmental conditions.

Having acknowledged the difficulties involved in equating the units of experimental taxonomy with those of orthodox taxonomy and indeed, the inadvisability of attempting to do so, the relationship of certain units of the two classifications have been briefly reviewed and it must be clear that at least some of the difficulties arise from the differing interpretations which have been given to the various terms employed. Of the units of experimental taxonomy, the concept of the coenospecies and that of the ecospecies have been fairly generally agreed upon but the concept of the ecotype has been variously interpreted. Lawrence (1945) said "it is ---- worthy of emphasis that the concept of the ecotype must be completely divorced from the morphological concept of species, subspecies or other taxonomic categories" but a degree of overlapping is inevitable when, as has been seen, the ecotype may coincide with, for example, the taxonomic subspecies. Nevertheless, it is all important that the concepts should remain distinct and it is, therefore, vital that the interpretation of the concepts should be clear and as far as possible, widely accepted. Since so far this remains an ideal, it is necessary to examine the ways in which the term ecotype has been used.

Lawrence himself through experimental methods, distinguished five ecotypes within Deschampsia caespitosa ssp. genuina. He found that clonal samples from five widely separated habitats from a climatic transect of California together with population samples from Finland, Swedish Lappland and South Sweden, retained their peculiar characteristics when grown together under a uniform environment whether the altitude was 100 feet, 4600 feet or 10,000 feet. Furthermore, each population reached its fullest development at that trial ground where conditions most nearly approximated to the original habitat, thus establishing that each represented a distinct ecological population. In applying the term ecotype to these five populations which are morphologically similar but physiologically adapted to different environments, Lawrence has followed closely the definition first given by Turesson (1922) of the ecotype as "the product arising as a result of the genotypical response of an ecospecies to a particular habitat." Clausen, Keck and Hiesey (1940), on the other hand, also followed Turesson's definition closely in delimiting ecotypes in Potentilla glandulosa but the four major and two minor ecotypes are all taxonomically recognised as subspecies. These authors also suspect the existence of two further ecotypes which are not morphologically differentiated, within two of the subspecies. Kovalev (1939) seems to have interpreted Turesson's definition very broadly in his delimitation of ecotypes within the cherry plum, Prunus cerasifera Ehrh. from a very wide distribution area. His ecotypes are in most cases named after the geographical regions in which they occur and since it is not stated that they have been grown experimentally under uniform conditions, the evidence concerning various attributes such as drought resistance, earliness of flowering etc. must be regarded with some dubiety. In any case the main emphasis has been upon morphological details and each ecotype has also been given specific or subspecific rank which makes it appear that the author has taken for granted that the populations must be ecotypes since they are morphologically differentiated and grow under different environmental conditions. If this is, indeed, the basis of classification as an ecotype, such an inexact application of the term must be deplored.

The ecotypes in Deschampsia caespitosa ssp. genuina and those in Potentilla glandulosa have this in common that they are separated from each other by a spatial interval even though as in the latter case, there is intergradation between them. Clausen et al. also note, however, that "each of the regional ecotypes contains a complex of biotypes" and give the example of P. glandulosa ssp. reflexa from Tuolumne Canyon. This is the foothill ecotype but it extends to relatively high levels in this particular canyon and when plants from these higher levels are grown under uniform conditions with plants from lower levels of the same canyon, they show a more dwarf habit of growth though there is no difference in earliness. The authors thus recognise ecotypic differentiation within the ecotype though they do not attempt to examine it in detail. As has already been noted, Sinskaja goes further in introducing the concept of the ecoelement but the concept of an internal cline related to a particular environmental factor would seem to be the most universally applicable and most generally informative when dealing with such variation. Gregor (1944) comes to the conclusion that as the ecotype as recorded by Clausen et al., and many other writers, really denotes "those populations which occupy a particular range of ecotypic variation on an ecocline"it ought preferably to be called an <u>ecoclinal subspecies</u> and suggested that <u>ecoclinal ecotype</u> "should be regarded as a subjective category of a subsidiary and complementary scheme of classification designed to record the general trends of ecotypic differentiation." This proposal would restrict the application of "ecotype" to ranges of character variation and would make it a more precise term, more finely adjusted to Turesson's original definition. There is much to be said for such an interpretation, the only objection perhaps being found in the use of "subspecies" even though prefixed by "ecoclinal" since this term is so well known though variously interpreted, taxonomically. Huxley (1942, p. 406) however, suggested that while subspecies have usually been defined with a geographical basis, there is no reason why other bases should not be employed as long as the particular basis is made clear by, for example, prefixing G for geographical, E for ecological and so on. The value of this interpretation of the term ecotype is obvious in the case of <u>Plantago maritima</u> where Gregor (1938) delimited three ecotypes, <u>decumbens</u>, <u>ascendens</u> and <u>erecta</u>, within what appeared to be a population containing a confused number of variate biotypes. Arranging the colonies on an edaphic basis grading from water-logged mud to relatively fertile coastal meadows, Gregor pointed out that each ecotype is represented in each edaphic habitat but represented in different proportions, <u>decumbens</u> becoming less frequent, <u>erecta</u> more frequent as the gradient passes to edaphically better conditions. It is interesting to note the similarity of these ideas to those later put forward by Sinskaja (1942) and it seems most probable that she would hold that these ecoclinal ecotypes are analagous with her concept of the ecoelement. It seems unnecessary, however, to coin a new term when the familiar ecotype may be used.

Sinskaja emphasis the need for examining whole "morphological and biological constitutions" but while this is certainly the ideal, it is impossible of fulfilment not only on account of lack of adequate knowledge, a stumbling block which may in time be overcome, but on account of the length of time involved and, therefore, the changes which may take place in the constitutions before an investigation could be completed. It is, however, true to say that some factors are of more importance than others, some aspects of the life of the organism have a greater effect upon its survival or evolution. Therefore, after careful consideration certain attributes may be found to be particularly valuable as <u>indicators</u> of the whole constitution. It is, nevertheless, useless to examine individual indicator attributes or variation in general except in relation to the primary factors which initiate the differentiation. Gregor (1938) for instance, realised that habit of growth was an important indicator attribute of the sea plantains from the edaphic point of view although if considered by itself this character could not indicate anything but the presence of variates in a population. Regarded in conjunction with the edaphic conditions, the true significance becomes apparent and the value of growth habit as an indicator of constitutional change in relation to environment may be appreciated.

With reference to the distribution of variation in the fescue populations (F. ovina and F. vivipara) of the Ben Cruachan area, it would seem likely that the most satisfactory interpretation may be obtained by an examination of the material in the light of these conclusions. It is obvious that no ecoclinal subspecies can be delimited but a pattern of differentiation can be traced. Omitting for the present the polyploid population of the sea-level area, it is possible to state that the fescue population of Ben Cruachan can be little affected by edaphic conditions since they are comparatively uniform throughout. Geographical isolation in the usual sense is non-existent and the small degree of spatial isolation is largely due to the phytosocial environment. There is, however, a definite change in climatic conditions with increase in altitude and since the statistical results prove that differences in certain attributes are correlated with altitude, it must be obvious that the climatic factor exerts a powerful selective influence. Of the various characters which have shown a definite gradient of change correlated with increasing height, the most easily observed at all times of the year is the habit of growth. The total height of the tallest flowering stem does not prove to be such a useful indicator as it is difficult to estimate on account of the general laxness of the plants. If, then, habit of growth is taken as an indicator attribute, it is possible to define three ecological ecotypes, decumbens, ascendens and erecta on a climatic ecocline. The unit populations at the lower levels are almost entirely formed from *erecta* whilst those at and near the summit consist almost exclusively of *decumbens* and the intermediate stations show a gradual change in the frequency of the occurrence of both. It ought, perhaps, to be emphasised here that the term ecotype can only be applied to a population not to an individual and, therefore, there can be no confusion between the ecotype *decumbens* for example, and what might be called "forma *prostrate*" by the orthodox taxonomist. As with the habit of growth and plant size, the lamina length and fertility of the populations decrease with altitude whilst leaf breadth and earliness increase so that ecoclines related to the climatic conditions may also be defined in respect of these attributes.

These gradual changes correlated to increase of altitude are distinct only from about 1,400 feet. At the lower levels an impression rather of a random distribution of variation is obtained and in the discussion of the results this was attributed to the phytosocial environment. Thus the populations are not continuous although the spatial distance would not appear to be sufficient to prevent cross-pollination. Bateman (1947), however, has shown that contamination in Maize falls sharply within the first ten to twenty feet and that at a distance of sixty feet, it has been reduced to 1%. Experiments by Jensen and Bøgh (1941) with various crops (quoted by Bateman) have given very similar graphical curves for ryegrass and cocksfoot although the distances involved are increased owing to the greater buoyancy of the pollen. Nevertheless, "there is an appearance of a minimal value beyond 500-600 m. of about one-twentieth the concentration (of pollen) at the source." Further, Gregor (1946) found that "in the wind-pollinated sea-plantain a spatial separation well within the dispersal limit of pollen is apparently sufficient to afford a degree of isolation which will allow communities to assume a genetic individuality of their own." If, then, a comparatively short distance can reduce cross-pollination to such an extent, the fescue populations have probably acquired part of their individuality through the process of genetic drift (Wright, 1940) since they are small in size. Spencer (1947) was able to present an excellent illustration of the random variation within small populations from his investigations into the genetic constitutions of "village"populations of <u>Drosophila</u> <u>immigrans</u>. He found that in these small colonies of flies overwintering in favourable conditions and isolated from all other colonies, a mutant gene of no selective advantage could attain a high frequency in the following breeding season and, provided it was not actually harmful, could persist presumably for years. Different "village" colonies showed a prevalence of differing mutations according to the reservoir of potential variability in the flies which chanced to survive the severe winter. With plants, once a colony has become isolated, there is less possibilty of new additions to the reservoir of change and, therefore, even more likelihood of marked differences between colonies. Fisher and Ford (1947) have, however, emphasised the inadvisability of attaching undue significance to the evolutionary importance of chance mutations in small isolated communities. They have come to the conclusion as a result of experiments with the moth Panaxia dominula L., that "the observed fluctuations in gene-ratio are much greater than could be ascribed to random survival only" and postulate that natural selection which varies from time to time in direction and intensity, is a vital factor whatever the size of the population so that while the Sewall Wright effect is unquestioned, its ultimate importance from the point of view of evolution may not be as great as some authors have been inclined to believe.

The selective action of the environment may be traced in spite of the irregular nature of the distribution of variation in the fescue population at the lower levels since there is a tendency, somewhat ill-defined it must be admitted, for at least some of the character ecoclines to be reflected there also. It does not seem unreasonable to suggest that the effect of the spatial isolation caused by the phytosocial environment is greater at the lower levels than the influence of the climatic factor but that the latter is also effective.

IX. CONCLUSION.

Professor Huxley has said (1942, p.38), "the description of internal clines when such exist within subspecies or monotypic species will serve as a useful corrective to the false sense of regional uniformity conferred by a binomial or trinomial name." This is abundantly clear in the case of the Ben Cruachan fescue populations which the data have proved to be the reverse of uniform. It would neither be helpful nor yet in accordance with the facts to name the mountain population of Festuca ovina for example, F. ovina var. montana nor would an improvement be effected by designating F. ovina and F. vivipara as mountain ecotypes. Such treatment does not go far enough since it only serves to throw into relief the fact that the populations are capable of survival under mountain environmental conditions. More information would be given by a delimitation of low-level, mid-level, and high-level ecotypes since at least some ecotypic trend would be suggested within the areal population but such a delimitation would also suggest definite boundaries which do not in fact exist. None of these treatments of the fescue populations give any indication of the nature of the adaptation which may be observed nor is it possible to deduce therefrom the probable relationship with populations occupying other types of habitat. In short it is both futile and inaccurate to give any population ecotypic status without reference to the characteristics of at least one other population on the same ecological gradient.

A further objection to designating the areal population a mountain ecotype or, for that matter, dividing it into low-level, mid-level, and high-level ecotypes, terms that infer climatic influence, is that while it is quite plain from the measurement data that the distribution of much of the variation is closely related to climate, internal trends would not only be largely ignored but this classification would also preclude the recognition of variation relating to any other environmental influence or else the population would require to be named again in respect of, for example, its biotic relationships. The first alternative is partial and unsatisfactory while the second is both unorthodox and plainly absurd.

The total ecotypic variation within a population will never be found to be related to a single environmental influence though one factor in the environment may well have a predominating effect, as climate has in the case of the Ben Cruachan fescues. The most satisfactory method of recording the variation, therefore, seems to be the application of the idea of a character gradient in its relation to the environmental factor which influences the character in question. In the present material, the definition of a climatic ecocline in respect of habit of growth not only acknowledges the existence of an internal trend of variation but points to the related agent in the environment and also indicates the most obvious evidence of hereditary adaptation. The choice of an indicator attribute is not an easy matter especially where a number of characters are related to the same environmental factor, but habit of growth is readily recognised and the variation can be appreciated even without recourse to measurement although in the present case, eye observations were confirmed by more exact methods.

It may be objected that in defining clines in terms of a single character, the total population individuality is in danger of being obscured in what might become an unwieldy mass of information. To a certain extent this is true but the advantages of this method are shown in the higher degree of accuracy which it provides and the greater possibilities it affords for understanding the relationship between population and environment. The total combination of population characteristics cannot at present be known so that the total which can be ascertained would still be less than the whole biological and morphological constitution demanded in theory, but not actually used, by Sinskaja. Even if it were possible, however, it would remain better to use indicator characters since all attributes are not influenced equally by any one environmental factor and character gradients do not necessarily follow parallel courses and, indeed, may even cross each other at right angles. Therefore, any attempt to record variation in terms of the total distinguishing attributes of climatically differentiated populations, as for instance, in the fescues, would result in confusion and would obscure even the primary response to climate. Since trends of variation in one character but related to different environmental gradients may cross each other, the population at the point of intersection will contribute data to both ecoclines. This does not mean, however, that the population is named more than once but only that the complex pattern of the distribution of variation has been adequately analysed and understood in its relationship to the environment.

It may be said that to split up the ecocline into ecoclinal ecotypes is inadvisable since the giving of a definite name tends to suggest more discontinuity than in fact exists; but it must be emphasised that an ecoclinal ecotype erecta, for example, is defined with regard to the relatively high frequency of erect types in the population. In the case of an edaphic ecocline, the arrangement may be scattered according to the irregular occurrence of the soil conditions and while this does not prevent the definition of an ecocline, it renders the division of it into ecotypes for reference purposes desirable. A climatic ecocline is naturally more regular, at least where the change in climate is gradual, but the advantage of defining ecotypes is not thereby lessened since reference to comparable populations on similar gradients is facilitated.

The details of the variation within populations on similar gradients are not necessarily identical, and indeed, an exact duplicate of any ecocline is unlikely if only because the genetic constitution of distantly separated populations may very probably be initially different. Again, the other environmental factors will scarcely bear the same relationship to each other and to climate in every case. The data obtained from the Ben Cruachan fescues show tht even two slopes of the same mountain do not provide exactly identical populations at the same altitude although the general trend of variation follows the same pattern. These differences must almost inevitably be increased by greater geographical separation so that the necessity for mentioning the location of ecoclines is made obvious if comparisons are to be made.

The regularity of the distribution of variation will be effected by the size and degree of isolation of the populations for random changes in gene frequency will tend to confuse the general picture where populations are small and more or less isolated. A further cause of irregularity is to be found in the fact that the selective pressure of, for example, the climatic conditions is unlikely to remain constant throughout the gradient. The detail of the distribution of variation will be affected but still the general trend will remain the same.

In conclusion it must be emphasised once more that there is no question of attempting to supersede the existing classifications of orthodox taxonomy nor yet of incorporating therein the data from experiment investigations. The genecological data can be used, however, to form an independent classification capable of providing a true picture of the variation, its distribution and relationship with the environment, particularly at the infra-specific level. Such a classification, subsidiary though it may be for general purposes, enables more precise comparisons to be made between populations under analagous conditions and also makes a considerable contribution to the ultimate understanding of evolutionary processes. Descriptive ecology it is true is content to employ the taxonomic species as its unit of comparison and for general survey work this method has proved its usefulness. The ecologist, however, is not as a rule, concerned with the details of the structure of specific tolerance to environment and it is precisely at this point that the genecological approach opens up a new and interesting field of study. This, too, is a subject not only of great academic importance but one which is capable of supplying data of economic value particularly where plants of agricultural use are under consideration.

X. SUMMARY

1. The investigation of the chromosome numbers of the fescue populations on Ben Cruachan shows that there are several distinct chromosome races. The setaceous-leaved sexual fescues are almost entirely diploid but a tetraploid also occurs: the broad-leaved sexual fescues include hexaploid and octoploid races as well as some heptaploids: the viviparous fescues are without exception tetraploid.

2. The viviparous fescues may be found throughout the area and occur all along each of the three gradients. In the setaceous-leaved sexual fescue the tetraploid occurs at sea-level but the diploid race extends as high as 2,600 feet on Ben Cruachan. The hexaploid race of broad-leaved sexual fescues has a range coincident with that of the sexual diploid although its occurrence on the hill is rather sporadic and the plants are rather difficult to identify. The octoploid and heptaploid races of the broad-leaved sexual fescues occur only at sea-level.

3. Chromosome counts of setaceous-leaved sexual fescues from various parts of Scotland, the Scottish islands, England and Wales indicate that the two chromosome races have a definite pattern of geographical distribution. The diploids occur in the North and islands whilst tetraploids are found in the South of Scotland, England and Wales but further investigations are necessary before any definite conclusions can be drawn.

4. The setaceous-leaved sexual fescues have been found to be capable of free interbreeding but are sexually distinct from the broad-leaved sexual fescues.

5. The hereditary variation which is present in each population makes it difficult to fit the plants into taxonomic groups but for reference purposes the setaceous-leaved sexual fescues are called Festuca ovina, the broad-leaved sexual fescues, Festuca rubra, and the viviparous fescues, Festuca vivipara.

6. A key for the identificanton of British viviparous fescues adapted by Wilmott from that proposed by Turesson is given in full and its application to the viviparous fescues from Ben Cruachan discussed.

7. The distribution of hereditary variation in F. ovina, F. rubra, and F. vivipara on the three gradients is discussed in the light of the statistical results obtained from the measurement data and an attempt is made to deduce reasonable explanations through correlation with environmental conditions.

8. The problem of the origin of hereditary variation in the viviparous fescues is discussed: mention is made of the occurrence of partial and complete sexual flowers on hunger inflorescences late in the season and also of the production of completely sexual plants from the bulbils of a viviparous plant.

9. The taxonomic units of species, subspecies and variety are discussed with special reference to the possibility and advisability of equating them with the units of experimental taxonomy.

10. The numerous difficulties involved in taking into account the hereditary variation in populations can only be adequately resolved by applying the concept of the character gradient with reference to the related environmental influence.

11. The areal ecotype is a useful but limited concept since the information which it supplies is little more than that the population in question is tolerant of the ecological conditions. The ecocline, however, takes into consideration the trends of hereditary variation which occur along an environmental gradient, and ecoclinal ecotypes may be defined for reference purposes. It is understandable that where a number of areal populations are investigated they may be found to occur on an environmental gradient and they may in that case be called ecoclinal subspecies. Such a classification permits of further definition within each ecoclinal subspecies of internal trends of variation.

12. The distribution of the hereditary variation within the fescue populations (F. ovina and F. vivipara) of Ben Cruachan in respect of habit of growth may be defined as following a climatic gradient. In this case, three ecoclinal ecotypes have been named for reference purposes, erecta, ascendens, and decumbens. The climatic ecocline in respect of habit of growth is quite plain in spite of some irregularity at the lower levels, an irregularity which in part, at any rate, is probably due to the small size and comparative isolation of the populations. Climatic ecoclines in respect of several other attributes may also be traced.

13. The chromosome races within F. rubra which occur at sea-level may be found to have distinct habitat preferences but no definite statement can be made until further research which is now being carried out, has been completed.

14. A review of the situation makes it clear that the units of orthodox taxonomy and those of experimental taxonomy must remain distinct. The classification of orthodox taxonomy has proved its usefulness and will continue to do so but the knowledge gained through the genecological approach will supplement it in a most valuable manner.

15. The study of trends of variation is not only of very great academic interest but it can also provide important data which are vital for the work of the plant breeder and for the economic success of agricultural crops.

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APPENDIX

Statistical Methods.

CALCULATIONS

CASE I. Let us consider a 7 x 5 area

	BLOCKS					TOTALS
Treatments	x_{11}	x_{12}	x_{13}	x_{14}	x_{15}	$x_{11}+x_{12}+x_{13}+x_{14}+x_{15}$ $= Y_1$
	x_{21}	x_{22}	x_{23}	x_{24}	x_{25}	$x_{21}+x_{22}+x_{23}+x_{24}+x_{25}$ $= Y_2$
	x_{31}	x_{32}	x_{33}	x_{34}	x_{35}	$x_{31}+\ldots\ldots\ldots+x_{35}$ $= Y_3$
	x_{41}	x_{42}	x_{43}	x_{44}	x_{45}	$x_{41}+\ldots\ldots\ldots+x_{45}$ $= Y_4$
	x_{51}	x_{52}	x_{53}	x_{54}	x_{55}	$x_{51}+\ldots\ldots\ldots+x_{55}$ $= Y_5$
	x_{61}	x_{62}	x_{63}	x_{64}	x_{65}	$x_{61}+\ldots\ldots\ldots+x_{65}$ $= Y_6$
	x_{71}	x_{72}	x_{73}	x_{74}	x_{75}	$x_{71}+\ldots\ldots\ldots+x_{75}$ $= Y_7$
Totals	x_{11} $+x_{21}$ $+x_{31}$ $+x_{41}$ $+x_{51}$ $+x_{61}$ $+x_{71}$ $=X_1$	x_{12} $+x_{22}$ $+$ $+$ $+$ $+$ $-x_{72}$ $=X_2$	x_{13} $+x_{23}$ $+x_{73}$ $=X_3$	x_{14} $+x_{24}$ $+$ $+$ $+$ $+$ $+x_{74}$ $=X_4$	x_{15} x_{25} $+$ $+$ $+$ $+$ $+x_{75}$ $=X_5$	$x_{11}+x_{12}+x_{13}+x_{14}+x_{15}$ $+\ldots\ldots$ to 35 terms = Grand Total $= X$

We shall assume that there is ONE plant in each plot.

1. Sum of squares

a) Blocks

$$\frac{1}{7}(X_1^2+X_2^2+X_3^2+X_4^2+X_5^2) - \frac{X^2}{35}$$

b) Treatments

$$\frac{1}{5}(Y_1^2+Y_2^2+Y_3^2+Y_4^2+Y_5^2+Y_6^2+Y_7^2) - \frac{X^2}{35}$$

c) Total

$$x_{11}^2+x_{12}^2+x_{13}^2+ \text{ .... to 35 terms} - \frac{X^2}{35}$$

Let B denote Block
T " Treatment
BT " Interaction of Block and Treatment. The interaction BT sum of squares is obtained by subtracting (a) + (b) from (c).

Analysis of Variance

(1) Variation due to	(2) Degrees of freedom	(3) Sum of Squares	(4) Variances	(5) Ratio of Variance
B	4	(a)	$\frac{(a)}{4}=(A)$	$\frac{(A)}{(C)}$
T	6	(b)	$\frac{(b)}{6}=(B)$	$\frac{(B)}{(C)}$
BT	24	(c)- (a)+(b)	$\frac{(c)-(a)+(6)}{24}=(C)$	
Total	34	(c)		

Apply F-test to ratio in column (5)

CASE II.

One or more plots may have no record Apply Missing Plot Technique to above 7 x 5 experiment in which we shall assume that x_{25}, x_{43}, x_{63} say are missing.

Denote missing values by a, b, c.

We then have the following table:-

						Totals.
	x_{11}	x_{12}	x_{13}	x_{14}	x_{15}	$x_{11}+x_{12}+x_{13}+x_{14}+x_{15}$
	x_{21}	x_{22}	x_{23}	x_{24}	a	$x_{21}+x_{22}+x_{23}+x_{24}+a$
	x_{31}	x_{32}	x_{33}	x_{34}	x_{35}	$x_{31}+x_{32}+x_{33}+x_{34}+x_{35}$
	x_{41}	x_{42}	b	x_{44}	x_{45}	$x_{41}+x_{42}+b+x_{44}+x_{45}$
	x_{51}	x_{52}	x_{53}	x_{54}	x_{55}	$x_{51}+x_{52}+x_{53}+x_{54}+x_{55}$
	x_{61}	x_{62}	c	x_{64}	x_{65}	$x_{61}+x_{62}+c+x_{64}+x_{65}$
	x_{71}	x_{72}	x_{73}	x_{74}	x_{75}	$x_{71}+x_{72}+x_{73}+x_{74}+x_{75}$
						Grand Total
	x_{11}	x_{12}	x_{13}	x_{14}	x_{15}	$x_{11}+x_{12}+x_{13}+x_{14}+x_{15}$
	$+x_{21}$	$+x_{22}$	$+x_{23}$	$+24$	$+a$	$+x_{21}+$ .... $+x_{24}+a$
	$+x_{31}$	$+x_{32}$	$+x_{33}$	x_{34}	$+x_{35}$	$+x_{31}+$ .... $+x_{35}$
Totals	$+x_{41}$	$+x_{42}$	$+b$	$+x_{44}$	$+x_{45}$	$+x_{41}+x_{42}+b+x_{44}+x_{45}$
	$+x_{51}$	$+x_{52}$	$+x_{53}$	$+x_{54}$	$+x_{55}$	$+x_{51}+$ .... $+x_{55}$
	$+x_{61}$	$+x_{62}$	$+c$	$+x_{64}$	$+x_{65}$	$+x_{61}+x_{62}+c+x_{64}+x_{65}$
	$+x_{71}$	$+x_{72}$	$+x_{73}$	$+x_{74}$	$+x_{75}$	$+x_{71}+$ .... $+x_{75}$

<u>Sums of Squares</u> <u>excluding</u> terms not necessary to finding of values for <u>a</u>, <u>b</u>, <u>c</u>,

a) Blocks

$$\tfrac{1}{7}\,(x_{13}+x_{23}+x_{33}+b+x_{53}+c+x_{73})^2+(x_{15}+a+x_{35}+x_{45}+x_{55}+x_{65}+x_{75})^2$$

$$-\tfrac{1}{35}\left\{\begin{array}{l} x_{11}+x_{12}+x_{13}+x_{14}+x_{15} \\ +x_{21}+x_{22}+x_{23}+x_{24}+a \\ +x_{31}+x_{32}+x_{33}+x_{34}+x_{35} \\ +x_{41}+x_{42}+b+x_{44}+x_{45} \\ +x_{51}+x_{52}+x_{53}+x_{54}+x_{55} \\ +x_{61}+x_{62}+c+x_{64}+x_{65} \\ +x_{71}+x_{72}+x_{73}+x_{74}+x_{75} \end{array}\right\}^2$$

b) Treatments

$$\tfrac{1}{5}\ (x_{21}+x_{22}+x_{23}+x_{24}+a)^2+(x_{41}+x_{42}+b+x_{44}+x_{45})^2+(x_{61}+x_{62}+c+x_{64}+x_{65})^2$$

$$\tfrac{1}{35}(x_{11}+x_{12}+\ \ldots\ldots\ \text{to 35 terms as shown in (a) above})^2$$

c) Total

$$a^2+b^2+c^2-\tfrac{1}{35}(x_{11}+x_{12}+\ldots\ \text{to 35 terms as shown in (a) above})^2$$

d) Form Residual Sum of Squares by subtracting (a)+(b) from (c) that is

$$a^2+b^2+c^2-\tfrac{1}{35}(x_{11}+x_{12}+\ \ldots\ \text{to 35 terms})^2$$

$$-\tfrac{1}{7}\ (x_{13}+x_{23}+x_{33}+b+x_{53}+c+x_{73})^2+(x_{15}+a+x_{35}+x_{45}+x_{55}+x_{65}+x_{75})^2$$

$$+\tfrac{1}{35}(x_{11}+x_{12}+\ \ldots\ldots\ \text{to 35 terms})^2$$

$$-\tfrac{1}{5}\ (x_{21}+x_{22}+x_{23}+x_{24}+a)^2+(x_{41}+x_{42}+b+x_{44}+x_{45})^2+(x_{61}+x_{62}+c+x_{64}+x_{65})^2$$

$$+\tfrac{1}{35}(x_{11}+x_{12}+\ \ldots\ldots\ \text{to 35 terms})^2$$

$$= a^2+b^2+c^2-\tfrac{1}{7}\ (x_{13}+x_{23}+x_{33}+b+x_{53}+c+x_{73})^2+(x_{15}+a+x_{35}+x_{45}+x_{55}+x_{65}+x_{75})^2$$

$$-\tfrac{1}{5}\ (x_{21}+x_{22}+x_{23}+x_{24}+a)^2+(x_{41}+x_{42}+b+x_{44}+x_{45})^2+(x_{61}+x_{62}+c+x_{64}+x_{65})^2$$

$$+\tfrac{1}{35}(x_{11}+x_{12}+\ \ldots\ldots+\ \text{to 35 terms})^2$$

Make the Residual Sum of Squares a minimum by differentiating with respect to <u>a</u>, <u>b</u>, <u>c</u>. This gives three equations which can be solved for <u>a</u>, <u>b</u>, <u>c</u>.
The equations are as follows:-
Differentiate with respect to <u>a</u>

Then

$$a + \tfrac{1}{35}\left\{\begin{array}{l} x_{11}+x_{12}+x_{13}+x_{14}+x_{15} \\ +x_{21}+x_{22}+x_{23}+x_{24} \\ +x_{31}+x_{32}+x_{33}+x_{34}+x_{35} \\ +x_{41}+x_{42}+x_{43}+x_{44}+x_{45} \\ +x_{51}+x_{52}+x_{53}+x_{54}+x_{55} \\ +x_{61}+x_{62}+x_{64}+x_{65} \\ +x_{71}+x_{72}+x_{73}+x_{74}+x_{75} \\ +a+b+c \end{array}\right\}$$

Continued over page

$- \frac{1}{7}(x_{15} + a + x_{35} + x_{45} + x_{55} + x_{65} + x_{75})$

$- \frac{1}{5}(x_{21} + x_{22} + x_{23} + x_{24} + a) \qquad = 0$

Differentiate with respect to <u>b</u>

$b + \frac{1}{5}$ Same as in above equation $- \frac{1}{7}(x_{13}+x_{23}+x_{33}+b+x_{53}+c+x_{73})$

$- \frac{1}{5}(x_{41} + x_{42} + x_{43} + x_{44} + b) \qquad = 0$

Differentiate with respect to <u>c</u>

$c + \frac{1}{35}$ Same as in above equation $- \frac{1}{7}(x_{13}+x_{23}+x_{33}+b+x_{53}+c+x_{73})$

$- \frac{1}{5}(x_{61} + x_{62} + x_{64} + x_{65} + c) \qquad = 0$

Having found <u>a</u>, <u>b</u>, <u>c</u> insert in table and proceed as in case I with the following degrees of freedom

	Degrees of freedom	Sum of Squares
B	4	
T	6	
BT	21	
Total	31	

Note:- There are 32 squares or plots in the original data, and thus the total number of freedoms is 32-1 = 31.

Example:-

Diploid June 1945

Leaf Length Mean Values one for each square

BLOCKS

Treatments

	A	B	C	D	E	F	TOTALS
30	100.75	147.75	129.25	148.75	135.67	a	a +662.17
37	113.67	112.00	120.25	b	126.50	147.00	b +619.42
5	155.00	119.75	145.00	136.50	175.75	119.75	851.75
6	102.50	93.50	115.75	124.75	132.00	116.00	684.50
7	133.75	119.75	111.00	92.50	104.75	136.00	697.75
27	238.00	c	165.00	150.00	194.25	156.00	c +903.25
40	188.50	124.00	d	160.00	167.00	147.00	d +786.50
14	160.50	133.33	122.25	135.25	190.50	123.50	865.33
25	129.25	99.25	141.25	120.50	112.75	139.75	742.75
24	127.00	118.75	121.00	114.75	116.25	119.25	717.00
23	120.00	102.50	105.00	111.00	130.25	115.25	684.00
22	90.75	87.00	108.75	83.75	110.25	76.00	556.50
TOTALS	1659.67	1257.58 + c	1384.50 + d	1377.75 + b	1695.92	1395.50 + a	8770.92 + a + b + c + d = Grand Total

Residual Sum of Squares (neglecting unnecessary numerical terms)

$$= a + b + c + d + {}^{1}/_{72}(8770.92 + a + b + c + d)^2$$

$$- {}^{1}/_{12}\ (1395.50 + a)^2 + (1377.75 + b)^2 + (1257.58 + c)^2 + (1384.50 + d)^2$$

$$- {}^{1}/_{6}\ (662.17 + a)^2 + (619.42 + b)^2 + (903.25 + c)^2 + (786.50 + d)^2$$

Differentiate with respect to <u>a</u>.

Then

$$a + {}^{1}/_{72}(8770.92+a+b+c+d) - {}^{1}/_{12}(1395.50+a) - {}^{1}/_{6}(662.17+a) = 0$$

i.e.

$$\begin{array}{llll} & a + 0.013889b + 0.013889c + 0.013889d & & + 121.818 \\ + & 0.013889a & & - 116.292 \\ - & 0.083333a & & - 110.362 \\ - & 0.016667a & = 0 & \end{array}$$

i.e.

$$0.763889a + 0.013889b + 0.013889c + 0.013889d = 104.836 \quad \ldots(1)$$

Similarly

$$0.013889a + 0.763889b + 0.013889c + 0.013889d = 96.231 \quad \ldots(2)$$

$$0.013899a + 0.013889b + 0.763889c + 0.013889d = 133.542 \quad \ldots(3)$$

$$0.013889a + 0.013889b + 0.013889c + 0.763889d = 124.640 \quad \ldots(4)$$

Let <u>b</u>, <u>c</u> and <u>d</u> have values $\frac{8770.92}{68} = 129.0$ approx.

Then equation (1) gives

$$.763889a + 5.375 = 104.836$$

i.e. $a = 130.2$

In equation (2) let $a = 130.2$, $b = c = 129.0$

Then

$$0.763889b + {}^{1}/_{72}(130.2) + {}^{1}/_{72}(129.0) + {}^{1}/_{72}(129.0) = 96.231$$

Hence

$$b = \begin{array}{r} 96.231 \\ - 1.808 \\ - 3.583 \end{array} \Big/ 0.763889 = 118.9$$

In equation (3) let $a = 130.2$, $b = 118.9$, $d = 129.0$

Then $0.763889c + {}^{1}/_{72}(130.2) + {}^{1}/_{72}(118.9) + {}^{1}/_{72}(129.0) = 133.542$

i.e. $c = 167.9$

Equation (4) gives

$$d = \frac{124.640 - 1.808 - 1.651 - 2.332}{0.763889} = 155.6$$

Now start again with equation (1)

where $b = 118.9$, $c = 167.9$, $d = 155.6$

Then $$a = \frac{104.836 - 1.651 - 2.332 - 2.161}{0.763889} = 129.20$$

In equation (2) let $a = 129.20$, $c = 167.9$, $d = 155.6$

Then $$b = \frac{96.231 - 2.332 - 2.161 - 1.794}{0.763889} = 117.74$$

In equation (3) let $a = 129.20$, $b = 117.74$, $d = 155.6$

Then $c = 167.50$

Then $d = 155.63$

Thus $a = 129.2$
$b = 117.7$
$c = 167.5$
$d = 155.6$

The original table has now been completed with BLOCK tables and TREATMENT totals as follows

	BLOCKS		TREATMENTS
A	1659.67		791.37
B	1425.08		737.12
C	1540.10		851.75
D	1495.45		684.50
E	1695.92		697.75
F	1524.70		1070.75
	9340.92	= Grand Total	942.10
			865.33
			742.75
			717.00
			684.00
			556.50
			9340.92

Sum of Squares

a) Blocks

$$(1659.67) + \ldots\ldots + (1524.70) - \frac{(9340.92)}{72}$$

$$= 4,431.02$$

b) Treatments $(791.37) + \ldots\ldots$ to 12 terms $- \frac{(9340.92)}{72}$

$$= 34,209.85$$

c) Total

$$(100.75)+(147.75)+(129.25)+(148.75)+(135.67)+(129.2)$$

$$+ \ldots\ldots \text{ to 72 terms } - \frac{(9340.92)}{72} = 55,628.18$$

<u>Table</u> Analysis of Variance
See Case II.

Variation due to	d.of f.	Sums of Squares	Variance	Variance Ratios
B	5	(a)	$\frac{(a)}{5}$ - (A)	$\frac{(A)}{(C)}$
T	11	(b)	$\frac{(b)}{11}$ - (B)	$\frac{(B)}{(C)}$
BT	51	$(c) - \{(a)+(b)\}$	$\frac{(c) - \{(a)+(b)\}}{51}$ -(C)	
Total	67	(c)		

Variation due to	d.of f.	Sums of Squares	Variance	Ratio of Variances
B	5	4,431.02	886.2 - (A)	$\frac{(A)}{(C)} = \overset{**}{2.66}$
T	11	34,209.85	3109.1 - (B)	$\frac{(B)}{(C)} = \overset{**}{9.33}$
BT	51	16,987.31	333.08-(C)	
Total	67	55,628.18		

** SIGNIFICANT AT 1%.

Case III. In this case each plot has 4 plants. As for example (using a similar notation to Case I).

BLOCKS					TOTALS
$x_{11}^{I.}$ $+x_{11}^{II.}$ $+x_{11}^{III.}$ $+x_{11}^{III.}$ $= x_{11}$	$x_{12}^{I.}$ $+x_{12}^{II.}$ $+x_{12}^{III.}$ $+x_{12}^{III.}$ $= x_{12}$				$x_{11}+x_{12}+x_{13}+x_{14}+x_{15} = Y_1$
$x_{21}^{I.} + x_{21}^{II}$ $+x_{21}^{III.} + x_{21}^{III.}$ $= x_{21}$	$x_{22}^{I} + x_{22}^{II}$ $+x_{22}^{III.} + x_{22}^{III.}$ $= x_{22}$				$x_{21}+x_{22}+x_{23}+x_{24}+x_{25} = Y_2$
x_{11} $+x_{21}$ $+x_{31}$ $+x_{41}$ $+x_{51}$ $+x_{61} = x_1$ $+x_{71}$					Grand Total $= x_1+x_2+x_3+x_4+x_5$ $= Y_1+Y_2+Y_3+Y_4+Y_5+Y_6+Y_7$

Sums of Squares

a) Blocks - similar to Case I.

viz. $1/28(x_1^2+x_2^2+x_3^2+x_4^2+x_5^2) - \frac{(x_1+x_2+x_3+x_4+x_5)^2}{140}$

b) Treatments = similar to Case I.

viz. $1/20(Y_1^2+Y_2^2+\ldots.+Y_7^2) - \frac{(x_1+x_2+x_3+x_4+x_5)^2}{140}$

c) Calculate

$$\frac{1}{4}\left\{\begin{array}{l} x_{11}+x_{12}+x_{13}+x_{14}+x_{15} \\ +x_{21}+x_{22}+x_{23}+x_{24}+x_{25} \\ +x_{31}+x_{32}+x_{33}+x_{34}+x_{35} \\ +x_{41}+x_{42}+x_{43}+x_{44}+x_{45} \\ +x_{51}+x_{52}+x_{53}+x_{54}+x_{55} \\ +x_{61}+x_{62}+x_{63}+x_{64}+x_{65} \\ +x_{71}+x_{72}+x_{73}+x_{74}+x_{75} \end{array}\right\} - \frac{(X_1+X_2+X_3+X_4+X_5)}{7 \times 5 \times 4}$$

This is equal to the sum of (a) and (b) and first order interaction BT. Hence to find sum of squares for BT subtract (a) + (b) from above sum (c) Total sum of squares

$$\left\{\begin{array}{l} x_{11}^{I^2} + x_{11}^{II^2} + x_{11}^{III^2} + x_{11}^{III^2} + \ldots \text{ to } (7 \times 5 \times 4) \text{ terms} \\ \text{i.e. to 140 terms} \end{array}\right\} - \frac{(X_1+X_2+X_3+X_4+X_5)^2}{140}$$

Analysis of Variance

Variation due to	d.of f.	Sums of squares	
B	4	(a)	
T	6	(b)	& c
BT	24	see above	
Residual	105		
Total	139		

Use F-test to test. BT is tested against Residual, B and T are tested against BT unless Residual variance is greater than BT variance.

Case IV. The number of plants in each plot are not equal; they may vary from 0 to 4.

continued/

First Method

Adjust number of plants in each square so that they are proportional. The main effects B, T and the first order interaction are calculated from the adjusted data, the residual being calculated from the original data. The adjusted numbers must not differ significantly from the original numbers.

Example

We shall first test whether the adjusted numbers are likely to differ significantly from the original numbers.

Leaf Length (diploid)
June 1945.

Number of plants in each plot with adjusted numbers in brackets.

	BLOCKS						
Treatments.	A	B	C	D	E	F	Total
30	4(3.48)	4(3.17)	4(3.48)	4(3.09)	3(3.32)	0(2.45)	19
37	3(3.30)	3(3.00)	4(3.30)	0(2.92)	4(3.15)	4(2.33)	18
5	4(4.40)	4(4.00)	4(4.40)	4(3.90)	4(4.20)	4(3.10)	24
6	4(3.85)	4(3.50)	4(3.85)	4(3.41)	4(3.68)	1(2.71)	21
7	4(4.03)	4(3.67)	4(4.03)	4(3.58)	4(3.85)	2(2.84)	22
27	1(2.57)	0(2.33)	4(2.57)	2(2.28)	4(2.45)	3(1.81)	14
40	4(1.47)	1(1.33)	0(1.47)	1(1.40)	1(1.40)	1(1.03)	8
14	4(3.67)	4(3.33)	3(3.67)	4(3.25)	2(3.50)	2(2.58)	20
25	4(4.40)	4(4.00)	4(4.40)	4(3.90)	4(4.20)	4(3.10)	24
24	4(4.40)	4(4.00)	4(4.40)	4(3.90)	4(4.20)	4(3.10)	24
23	4(4.40)	4(4.00)	4(4.40)	4(3.90)	4(4.20)	4(3.10)	24
Totals	44	40	44	39	42	31	240

In this example it will be noted that four plots contain no plants.

The adjusted numbers for treatment 30 are as follows:

$\frac{19}{240} \times 44$; $\frac{19}{240} \times 40$; $\frac{19}{240} \times 39$; $\frac{19}{240} \times 42$; $\frac{19}{240} \times 31$.

The sum of the terms $\frac{(\text{observed} - \text{calculated})^2}{\text{calculated}}$

one for each square gives χ^2
It is found that $\chi^2 = 23.81$ for 55^* degrees of freedom

This is not significant

*55 (5 x 11) entries can be made arbitrarily assuming the total figures for each row and column remain unaltered.

It can therefore be safely assumed that in all the other experiments the adjusted numbers will not differ significantly from the original numbers, since the example chosen was one in which greatest variation appeared.

We first consider the case where the number of plants in each square vary from 1 to 4.

Example

Leaf Length - Hexaploid
September, 1945.

The top figure is the mean length of plant for a square (obtained from original data)

The middle figure gives the original number of plants together with the adjusted number.

The bottom figure gives the square of the top figure.

(continued)/

	BLOCKS						Treatment Totals
eat- ts	440.8 4, 4.31 194,304.6	501.2 4, 4.21 251,201.4	340.0 4, 3.94 115,600.0	414.5 4, 3.94 171,810.2	523.2 4, 3.85 273,738.2	406.8 4, 3.76 165,486.2	10,526.518
	434.2 4, 4.31 188,529.6	419.8 4, 4.21 176,232.0	405.2 4, 3.94 164,187.0	311.2 4, 3.94 96,845.4	421.2 4, 3.85 177,409.4	418.2 4, 3.76 174,891.2	9,655.428
	468.2 4, 4.31 219,211.2	439.5 4, 4.21 193,160.2	446.0 4, 3.94 198,249.0	493.0 4, 3.94 185,072.0	430.2 4, 3.85 153,820.8	393.2 4, 3.76	10,699.339
	417.3 3, 2.15 174,139.3	365.5 2, 2.11 133,590.2	480.0 2, 1.97 230,400.0	270.0 1, 1.97 72,900.0	400.0 2, 1.92 160,000.0	519.5 2, 1.88 269,880.2	4,890.560
	387.8 4, 4.31 150,388.8	366.2 4, 4.21 134,102.4	395.0 4, 3.94 156,025.0	401.2 4, 3.94 160,961.4	454.8 4, 3.85 206,843.0	446.0 4, 3.76 198,916.0	9,778.088
	467.8 4, 4.31 218,836.8	307.2 4, 4.21 94,371.8	353.2 4, 3.94 124,750.2	500.0 4, 3.94 250,000.0	410.0 4, 3.85 168,100.0	431.5 4, 3.76 186,192.2	9,872.078
	388.0 4, 3.05 150,544.0	333.2 4, 2.98 111,022.2	396.3 3, 2.79 157,053.7	516.5 2, 2.79 266,772.2	360.0 2, 2.73 129,600.0	412.0 2, 2.66 169,744.0	6,801.768
	459.8 4, 4.31 211,416.0	377.8 4, 4.21 142,732.8	430.0 4, 3.94 184,900.0	498.5 4, 3.94 248,502.2	455.2 4, 3,85 207,207.0	467.2 4, 3.76 218,275.8	10,739.758
	407.5 4, 4.31 166,056.2	429.8 4, 4.21 184,728.0	448.8 4, 3.94 201,421.4	476.5 4, 3.94 227,052.2	444.8 4, 3.85 197,847.0	436.8 4, 3.76 190,794.2	10,566.310
	348.5 4, 4.31 121,452.2	376.5 4, 4.21 141,752.2	443.0 4, 3.94 196,249.0	428.0 4, 3.94 183,184.0	439.5 4, 3.85 193,160.2	409.5 4, 3.76 167,690.2	9,750.635
	456.2 4, 4.31 208,118.4	503.5 4, 4.21 253,512.2	521.5 4, 3.94 271,962.2	489.8 4, 3.94 239,904.0	530.2 4, 3.85 281,112.0	458.8 4, 3.76 210,497.4	11,836.837
	500.2 4, 3.05 250,200.0	420.5 4, 2.98 176,820.2	270.5 2, 2.79 73,170.2	461.0 4, 2.79 212,521.0	468.0 2, 2.73 219,024.0	431.0 1, 2.66 185,761.0	7,243.685
K ls	20,289.353	18,684.746	17,709.810	19,069.663	18,848.475	17,758.960	Grand Total 112,361.004

The Block totals are obtained by multiplying the top figure by the adjusted figure and adding for each column.

Thus the first BLOCK total

$$
\begin{aligned}
= \; & (440.8 \times 4.31)+(434.2)\,(4.31)+(468.2)\,(4.31) \\
& +(417.3 \times 2.15)+(387.8)\,(4.31)+(467.8)\,(4.31) \\
& +(388.0)\,(3.05)+(459.8)\,(4.31)+(407.5)\,(4.31) \\
& +(348.5)\,(4.31)+(456.2)\,(4.31)+(500.2)\,(3.05) \\
= \; & 20{,}289.353
\end{aligned}
$$

The BLOCK totals with the corresponding total adjusted number of plants are as follows:

20,289.353 , 47.04
18,684.746 , 45.96
17,709.810 , 43.01
19,069.663 , 43.01
18,848.475 , 42.03
17,758.960 , 41.04
112,361.007 , 262.09

Sum of Squares
<u>BLOCKS</u>

$$
\frac{(20{,}289.353)^2}{47.04} + \frac{(18{,}684.746)^2}{45.96} + \text{to 6 terms} - \frac{(112{,}361.004)^2}{262.09}
$$

$$= 61{,}647$$

The TREATMENT totals, obtained in a similar way, together with the corresponding adjusted number of plants for each treatment are as follows:-

10,526.518 , 24.01
9,655.428 , 24.01
10,699.399 , 24.01

(Continued)

4,890.560	,	12.00
9,778.088	,	24.01
9,872.078	,	24.01
6,801.768	,	17.00
10,739.758	,	24.01
10,566.310	,	24.01
9,750.635	,	24.01
11,836.837	,	24.01
7,243.685	,	17.00
112,361.004		262.09

Sum of Squares
TREATMENTS

$$\frac{(10,526.518)^2}{24.01} + \text{ - to 12 terms - } \frac{(112,361.004)^2}{262.09}$$

$$= 186,842$$

To find sum of squares for BT

We first, see Case 3, find B + T + BT.

It is given by using the bottom figure of a square multiplied by the adjusted number, adding for each square and subtracting $\frac{(112,361.004)^2}{262.09}$

That is (194,304.6) (4.31) + (251,201.4) (4.21) + ... to 72 terms

$$- \frac{(112,361.004)^2}{262.09}$$

$$= 885,477$$

Hence
Sum of Squares

$$BT = 885,447 - 61,647 - 186,842$$

$$= 636,958.$$

From the original data we find for the one-way classification into 72 squares that the
Sum of Squares = 745,608
and for the total from 262 plants
Sum of Squares = 2,054,078
We thus have the following table

September 11 Hexaploid 1945

Variation due to	d.off.	Sum of squares	Variance	Ratio of Variances	
B	5	61,647	(1) 12,329	$\frac{(1)}{(3)} = 1.06$	Adjusted data
T	11	186,842	(2) 16,986	$\frac{(2)}{(3)} = 1.47$	
BT	55	636,988	(3) 11,581.6	$\frac{(3)}{(4)} = 1.68$	
One-way classification	71	745,608			Original data
Residual	190	1,308,470	(4) 6,886.7		
Total		2,054,078			

We now consider the case where no measures of plants are available for a few of the squares, the remainder having 1 to 4 plants. The Missing Plot technique is applied, the mean value of the measures in any one square being used. The example previously given actually does use these mean values and it was found that the missing plots were given values

$a = 129$
$b = 118$
$c = 168$
$d = 156$

One plant was allotted to each missing square and we now proceed as in the previous example. In obtaining the adjusted numbers the original marginal totals were used, and see also note below on degrees of freedom.

Example Leaf Length (diploid) June 1945

The first table shows in any one square the original number of plants, assuming one plant in the "missing" squares; the total length for the number of plants and the sum of the squares of the lengths.

BLOCKS

TREATMENTS	A	B	C	D	E	F
30	4 403 41809	4 591 87499	4 517 68735	4 595 90993	3 407 56329	1* 129
37	3 341 39325	3 336 37694	4 481 58143	1* 118 14161	4 506 65626	4 588 87714
5	4 620 98186	4 479 60177	4 580 85816	4 546 75218	4 703 123951	4 479 57611
6	4 410 41264	4 374 39478	4 463 56997	4 499 63893	4 528 80782	1 116 13456
7	4 535 72219	4 479 60027	4 444 49804	4 370 35874	4 419 45193	2 272 37504
27	1 238 56644	1* 168	4 660 119262	2 300 45018	4 777 153887	3 468 77774
40	4 714 132710	1 124 15376	1* 154	1 160 25600	1 167 27889	1 147 21609
14	4 642 104210	4 533 71319	4 489 60423	4 541 76453	2 381 73581	2 247 33317
25	4 517 68085	4 397 40909	4 565 81519	4 482 65182	4 451 52737	4 559 79395
24	4 508 65308	4 475 59969	4 484 59194	4 459 55251	4 465 54861	4 477 57107
23	4 480 58092	4 410 42486	4 420 47388	4 444 51698	4 521 68749	4 461 52195
22	4 383 37351	4 348 30818	4 435 48331	4 335 28721	4 441 49811	2 152 11840

* Missing values inserted.

This table provides the data to determine the Residual variance (see below)

	d. of f.	Sums of squares	Variance
one-way classification	67	129,588	
Residual	172	112,747	655.5
Total	239	242,335	

Since the numbers of plants vary from 1 to 4 we employ the method detailed above, adjusting the frequencies so that they are proportional. The following table gives for each square

(1) The mean length of plant for a square (original data).

(2) The original and adjusted number of plants.

BLOCKS.

TREATMENTS

	A	B	C	D	E	F	Totals
30	100.75 4,3.48	147.75 4,3.17	129.25 4,3.48	148.75 4,3.09	135.67 3,3.32	129. (1)2.45	2494.9 18.99
37	113.67 3,3.30	112.0 3,3.00	120.25 4,3.30	118. (1)2.92	126.50 4,3.15	147.0 4,2.33	2193.5 18.00
5	155.0 4,4.40	119.75 4,4.00	145. 4,4.40	136.5 4,3.90	175.75 4,4.20	119.75 4,3.10	3440.8 24.00
6	102.5 4,3.85	93.5 4,3.50	115.75 4,3.85	124.75 4,3.41	132.0 4,3.68	116.0 1,2.71	2393.0 21.00
7	133.75 4,4.03	119.75 4,3.67	111.0 4,4.03	92.5 4,3.58	104.75 4,3.85	136.0 2,2.84	2546.5 22.00
27	238.0 1,2.57	168 (1)2.33	165.0 4,2.57	150.0 2,2.28	194.25 4,2.45	156.0 3,1.81	2527.4 14.01
40	178.5 4,1.47	124.0 1,1.33	154. (1)1.47	160.0 1,1.30	167.0 1,1.40	147.0 1,1.03	1246.9 8.00
14	160.50 4,3.67	133.33 4,3.33	122.25 4,3.67	135.25 4,3.25	190.50 2,3.50	123.50 2,2.58	2906.7 20.00
25	129.25 4,4.40	99.25 4,4.00	141.25 4,4.40	120.5 4,3.90	112.75 4,4.20	139.75 4,3.10	2964.0 24.00
24	127.0 4,4.40	118.75 4,4.00	121.0 4,4.40	114.75 4,3.90	116.25 4,4.20	119.25 4,3.10	2871.6 24.00
23	120.0 4,4.40	102.5 4,4.00	105.0 4,4.40	111.0 4,3.90	130.25 4,4.20	115.25 4,3.10	2737.2 24.00
22	95.75 4,4.03	87.0 4,3.67	108.75 4,4.03	83.75 4,3.58	110.25 4,3.85	76.0 2,2.84	2083.6 22.00
Totals	5845.8 44.0	4651.7 40.0	5530.8 44.0	4733.0 39.01	5786.0 42.00	3858.8 30.99	30406.1 240- Numbers.

BLOCK totals: (Calculation as before)

	P. Number
5845.8	44.0
4651.7	40.0
5530.8	44.0
4733.0	39.01
5786.0	42.00
3858.8	30.99
30406.1	240.0

Sum of squares

BLOCKS (B)

$$\frac{(5845.8)^2}{44.0} + \ldots\ldots\ \text{to 6 terms} - \frac{(30406.1)^2}{240.0}$$

$$= \begin{array}{r} 776,667.7 \\ 540,957.8 \\ 695,221.6 \\ 574,244.9 \\ 797,090.5 \\ 480,488.5 \end{array} - 3,852,212.2 = 12,458.8$$

TREATMENT totals

2494.9	18.99
2193.5	18.00
3440.8	24.00
2393.0	21.00
2546.5	22.00
2527.4	14.01
1246.9	8.00
2906.7	20.00
2964.0	24.00
2871.6	24.0
2083.6	22.00
2737.2	24.00
30406.1	240.00

Sum of squares
TREATMENTS (T)

$$\frac{(2494.9)^2}{18.99} + \text{to 12 terms} - \frac{(30406.1)^2}{240.0}$$

327,779.0
267,302.3
493,296.0
272,688.0
294,757.4
455,942.2 - 3,852,212.8
194,345.0
422,445.2
366,054.0
343,586.9
312,177.7
197,335.9
= 91,496.8

B + T + BT
Sum of squares

$$(100.75)^2\ (3.48) + (147.75)^2\ (3.17 + \ldots \text{to 72 terms} - \frac{(30406.1)^2}{240}$$

$$= \frac{(350.6)^2}{3.48} + \frac{(468.4)^2}{3.48} + \ldots\ldots\ldots\ldots\ldots\ldots \quad \text{do.} \quad \text{do.}$$

when 350.6 = (100.75) (3.48)
468.4 = (147.75) (3.17) + c

= 35,321.95 + 69,210.9 +58,137.9 + 68,359.9 + 61,102.5 + see below
+ 42,636.4 + 37,632.0 +47,712.2 + 40,667.5 + 50,413.4 +
+105,710.0 + 57,360.1 +92,510.0 + 72,679.4 +129,747.4 +
+ 40,443.9 + 30,588.5 +51,573.9 + 53,069.0 + 50,929.8 +
+ 72,089.6 + 52,632.2 +49,647.0 + 30,640.6 + 42,247.0 +
+145,594.1 + 65,748.5 +69,951.8 + 51.300.0 + 92,441.1 +
+ 46,839.3 + 20,445.1 +34,868.7 + 33,280.0 + 39,044.6 +
+ 94,528.9 + 59,200.0 +54,858.8 + 59,461.0 +127,034.9 +
+ 73,504.5 + 39,402.25+87,786.9 + 56,641.0 + 53,404.0 +
+ 70,967.6 + 56,406.25+64,420.4 + 51,347.7 + 56,747.4 +
+ 63,360.0 + 42,025.0 +48,510.0 + 48,051.9 + 54,573.6 +
+ 36,952.6 + 27,780.0 +47,669.2 + 25,106.2 + 46,805.2 +

+ 40,783.3
+ 50,346.0
+ 44,448.2
+ 36,475.0
+ 52,517.8 - 3,852,212.2
+ 44,060.6
+ 22,254.3 + 41,181.7
+ 39,343.4 + 16,397.8
+ 60,536.2
+ 44,089.8 = 3,981,555.7 = 129,343.5
- 3,852,212.2

Variation due to	d. of f.	Sum of squares	Variance	Ratios
B	5	12,458.8	(A) 2491.8	$\frac{(A)}{(D)}=3.80$
T	11	91,496.8	(B) 8318.0	$\frac{(B)}{(D)}=12.69$
BT	51	25,387.9	(C) 497.8	$\frac{(C)}{(D)}=0.76$
Residual	172	112,747	(D) 655.5	

These results may be compared with those using <u>one</u> mean value for each square, all squares being thus equally weighted. The example on p.9 whose data differs only slightly from the above, shows that, taking into consideration the differences in the number of degrees of freedom, it is possibly the more rigorous method. The gain from this point of view is, however, probably, more than offset by the loss of accuracy which is involved in taking one mean value for each plot since many plots only contain one value in the first place.

A rigorous method for unequal numbers plots has been given in Kendall "The Advanced Theory of Statistics," Volume II, pp. 220-226. We shall apply it to the Leaf Lengths-hexaploid September 1945. (see Case 4).

Class Means and Reciprocals of Class-Frequencies (see Case 4 for data).

							TOTALS
	440.8	501.2	340.0	414.5	523.2	406.8	2,626.5
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	434.2	419.8	405.2	311.2	421.2	418.2	2,409.8
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	468.2	439.5	446.0	493.0	430.2	392.2	2,669.1
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	417.3	365.5	480.0	270.0	400.0	519.5	2,452.3
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	387.8	366.2	395.0	401.2	454.8	446.0	2,451.0
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	467.8	307.2	353.2	500.0	410.0	431.5	2,469.7
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	388.0	333.2	396.3	516.5	360.0	412.0	2,406.0
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	459.8	377.8	430.0	498.5	455.2	467.2	2,688.5
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	407.5	429.8	448.8	476.5	444.8	436.8	2,644.2
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	348.5	376.5	443.0	428.0	439.5	409.5	2,445.0
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	456.2	503.5	521.5	489.8	530.2	458.8	2,960.0
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
	500.2	420.5	270.5	461.0	468.0	431.0	2,551.2
	.2500	.2500	.2500	.2500	.2500	.2500	1.5000
Totals	3.0833	3.2500	3.5833	4.0000	3.7500	4.2500	21.9166
	5176.3	4840.7	4929.5	5260.2	53337.1	5229.5	30773.3

q = 6

p = 12

<u>1st row</u> $\frac{1}{N_1} = \frac{1}{q^2}(.2500 + .2500 + .2500 + .2500 + .2500) = \frac{1.5}{36}$

TREATMENTS

		The unweighted marginal means	and squares
$N_1 = 24$	$N_1 = 24$	$\bar{\bar{x}}_1 = 437.8$	1916.69
$\frac{1}{N} = \frac{1.5}{36}$ ∴	$N_2 = 24$	$\bar{\bar{x}}_2 = 401.6$	161283.
	$N_3 = 24$	$\bar{\bar{x}}_3 = 444.9$	197936.
$\frac{1}{N_4} = \frac{3.3333}{36}$	$N_4 = 10.80$	$\bar{\bar{x}}_4 = 408.7$	167036.
$\frac{1}{N_5} = \frac{1.5}{36}$	$N_5 = 24$	$\bar{\bar{x}}_5 = 408.5$	166872.
	$N_6 = 24$	$\bar{\bar{x}}_6 = 411.6$	169415.
$\frac{1}{N_7} = \frac{2.3333}{36}$	$N_7 = 15.43$	$\bar{\bar{x}}_7 = 401.0$	160801.
	$N_8 = 24$	$\bar{\bar{x}}_8 = 448.1$	200793.
	$N_9 = 24$	$\bar{\bar{x}}_9 = 440.7$	194216.
	$N_{10} = 24$	$\bar{\bar{x}}_{10} = 440.8$	194305.
	$N_{11} = 24$	$\bar{\bar{x}}_{11} = 493.3$	243345.
$\frac{1}{N_{12}} = \frac{2.7500}{36}$	$N_{12} = 13.09$	$\bar{\bar{x}}_{12} = 425.2$	180795.

$$C = \frac{\sum N_j \bar{\bar{x}}_j}{\sum N_j} = \frac{24(437.8) + \ldots\ldots \text{ to 12 terms}}{255.32}$$

$$= \frac{110422.458}{255.32} = 432.49$$

Thus the estimate of variance with 11 degrees of freedom

$$= \frac{1}{11}\left\{\sum (N_j \bar{\bar{x}}_j^2) - C^2 \sum_j N_j\right\}$$

$$= \frac{1}{11}\left\{\begin{array}{r} 47,927,770.78 \\ -47,756,993. \\ \hline 170,778 \end{array}\right\} = 15,525$$

Variance for Treatments = 15,525

<u>1st Column</u>

$$\frac{1}{M_1} = \frac{1}{p^2}(3.0833) = \frac{1}{144}\quad(3.0833)$$

$$\begin{array}{llrr}
M_1 = 46.70 & \bar{x}_{\cdot 1} = 431.36 = & 431.4 & 186106. \\
M_2 = 44.31 & \bar{x}_{\cdot 2} = & 403.4 & 162732. \\
M_3 = 40.19 & \bar{x}_{\cdot 3} = & 410.8 & 168757. \\
M_4 = 36.00 & \bar{x}_{\cdot 4} = & 438.3 & 192107. \\
M_5 = 38.40 & \bar{x}_{\cdot 5} = & 444.8 & 197847. \\
M_6 = 33.88 & \bar{x}_{\cdot 6} = & 435.8 & 189922.
\end{array}$$

$$\underline{4361883.11}$$

$$c = \frac{102155.110}{239.48} = 426.57$$

Estimate of variance with 5 degrees of freedom

$$= \frac{1}{5}\left\{\begin{array}{r} 43{,}631{,}883. \\ \underline{-43{,}576{,}251.} \\ 55{,}632 \end{array}\right\} = 11{,}126$$

The Residual Variance is that given on p. 17.

Thus

Variation due to	d. of f.	Variance	Ratio
B	5	(A) 11,126	$\frac{(A)}{(C)} = 1.62$
T	11	(B) 15,525	$\frac{(B)}{(C)} = 2.25$
Residual	190	(C) 6,886.7	

This compares favourably with the approximate method (see page 17) and since the latter involves less labour in calculation, it was adopted for the data under review.