GENETIC VARIATION IN DIPLOID *DACTYLIS*

I. CROSS COMPATIBILITY AND SEEDLING CHARACTERS

P. F. PARKER

Hartley Botanical Laboratories, The University, Liverpool, 3

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1. INTRODUCTION

Dactylis is a small Eurasian genus of outbreeding perennials with a considerable number of morphologically and geographically distinct diploid ($2n = 14$) subspecies (Stebbins and Zohary, 1959; Parker and Borrill, 1968). Since the diploids and their hybrids are meiotically stable (Jones, 1962), the genetic basis of their variation is amenable to analysis, this has been carried out using the techniques developed by Jinks (1954), Hayman (1954*a, b*) and Dickinson and Jinks (1956) for diallel analysis.

Previous diallel studies in the Gramineae have been mainly concerned with fertility and seedling characters (Beddows, Breese and Cocks, 1962; Sun and Rees, 1963; Hayward and Breese, 1966). The present and following publications seek to study the mode of genetic differentiation occurring between populations of reproductively isolated but reasonably interfertile subspecies for a range of morphological and physiological characters in both seedling and adult stages.

2. MATERIALS AND METHODS

All populations used are morphologically representative samples of the geographically isolated subspecies considered. All seed samples were collected from native habitats (table 1), in each case the sample is based on several genotypes, although the exact number is unknown. The seed of each subspecies population was sown, and from the resultant plants, a sample of the required number was taken at random for crossing.

Self fertility of diploid *Dactylis* was studied by Borrill (1961), who showed that selfed plants were usually sterile. Where seedlings were produced, they were conspicuously reduced in vigour, and rarely reached maturity. In the light of these facts, also taking into account that the plants being crossed were very different from each other, and selfs easily identified, no further extensive tests were carried out.

To provide a parental population to compare with the hybrids, single panicles from each of twelve genotypes within a subspecies were enclosed in a glassine bag to interpollinate. Crossing between subspecies was based on random selection of plants from the different subspecies and pair crossing them, using the approach method of Jenkin (1924, 1931). Five pair crosses were completed between each of the subspecies, and the seed bulked after threshing, keeping cross and reciprocal separate.

Seed from the 72 inter-subspecies bulked crosses, and from the nine intra-subspecies bulked crosses, were sown in boxes in the glasshouse in autumn 1963, using a randomised block layout with three replications.

TABLE 1

Subspecific names, and origin of experimental material. The numbers in the left-hand column are those used to identify the subspecies in the W_r/V_r and $W'r/W_r$ graphs

No.	Subspecies	Location and Habitat	Distribution
1.	<i>Smithii</i>	La Orotava, Teneriffe. Shady crevices in volcanic rock. 150-800 m.	Canary Islands. Endemic.
2.	<i>junicinella</i>	Near Residencia Universitaria. Bare rocky slopes. 2000-3000 m.	Sierra Nevada, Spain. Endemic.
3.	<i>lusitanica</i>	Near Cintra, Portugal. Open woodland, scrubland, roadsides.	West and Central Portugal. Endemic.
4.	<i>aschersoniana</i>	Southern Sweden. Deciduous (Quercus-Fagus) forests.	Discontinuous, N.W. Europe to Caucasus.
5.	<i>parthiana</i>	Elburz Mts., Iran. Deciduous (Quercus-Fagus) forests.	Northern slopes of Elburz Mts., Iran.
6.	<i>Himalayensis</i>	Gangi, Tehri, India. Forest glades. 1800-4000 m.	Himalayas and W. China.
7.	<i>ibizensis</i>	San Antonio Abad. Open Maquis between forest and sea.	Islands of Ibiza and Formentera, Balearics. Endemic.
8.	<i>castellata</i>	Oued Chiffa. Railway embankment, and other disturbed habitats.	Algeria, and probably Morocco.
9.	<i>santai</i>	Relizane, Algeria. Open forest and garigue.	Algeria and W. Morocco.

Each of the 81 families was represented by a sowing of 24 seeds, later thinned to 12 plants.

The characters considered here are:

- (i) Heavy seed set.
- (ii) Heavy seed germination.
- (iii) Fertility index.
- (iv) Seedling growth rate. (The number of days from mean germination to mean leaf six-maturity.)
- (v) Seedling stem internode elongation. (Ground to base of leaf 9.)
- (vi) Leaf six area.
- (vii) Number of tillers at leaf six maturity (a leaf was judged mature when the leaf above was over half its length).

3. RESULTS

(i) Heavy seed set

As all hybridisation data were recorded as a single unreplicated mean figure, the usual analysis of variance is rather limited in application, all fertility data fall into the category of random sampling. Data for initial cross and F_1 are presented in table 2.

The analysis of variance devised by Hayman (1954*a*) was used (table 3); within this scheme there are two possible models to consider, one for maternal effects and one for reciprocal effects. With unreplicated data there is no

TABLE 2
Mean seed set of the initial crosses (upper line) and F_1 hybrids (lower line)

♂	♀ 1	2	3	4	5	6	7	8	9
1.	24.3 —	33.1 32.5	11.2 46.4	9.9 6.9	15.1 9.6	13.3 6.0	18.2 4.5	37.7 4.5	8.9 19.6
2.	25.1 57.4	41.7 —	23.4 24.0	35.4 13.5	8.0 14.0	12.2 5.8	22.8 6.6	38.8 25.4	20.7 6.4
3.	18.4 21.3	39.2 9.2	59.7 —	35.3 25.4	22.2 18.4	26.7 25.5	30.1 21.2	30.0 40.6	39.3 39.6
4.	12.5 8.9	47.0 6.9	52.0 19.3	25.0 —	33.0 19.0	27.3 3.1	23.2 8.8	35.1 3.7	35.7 4.1
5.	2.3 8.0	7.9 17.3	35.2 37.1	41.6 25.0	32.7 —	15.0 4.5	24.8 23.3	51.1 20.6	60.7 11.6
6.	6.2 10.0	30.4 1.2	51.7 27.2	31.8 2.9	16.1 3.7	33.7 —	16.8 5.9	28.9 18.0	35.3 2.6
7.	13.3 3.6	36.3 6.0	8.6 28.6	12.0 5.2	13.2 9.0	25.5 4.4	24.0 —	8.5 31.6	14.0 19.3
8.	15.8 5.4	51.1 22.9	25.5 28.2	27.9 2.7	20.4 10.3	16.4 8.3	14.6 45.4	40.3 —	27.0 23.3
9.	5.0 12.2	15.5 7.6	29.9 35.3	20.4 3.0	26.4 8.4	22.7 2.6	33.4 15.0	27.3 28.9	42.9 —

test of significance for specific reciprocal differences as measured by (*d*). When tested against (*d*) the items (*c*) measuring consistent reciprocal effects, (*b*) measuring dominance and epistasis, and (*a*) measuring additive gene action, are all significant in the initial cross, whereas in the F_1 there are no consistent reciprocal effects.

TABLE 3
Hayman's analysis of variance of unreplicated seed set data for both initial cross, and F_1 hybrids

Item	D.F.	Initial cross		F_1	
		M.S.	V.R.	M.S.	V.R.
<i>a</i>	8	517.41	7.68**	666.86	19.20***
<i>b</i>	36	169.91	2.52**	243.80	7.02***
<i>c</i>	8	216.47	3.21*	63.72	1.83
<i>d</i>	28	67.33	—	34.73	—

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

Further analysis of the data, using the graphical method originally developed by Jinks (1954) and Hayman (1954*b*) for inbred lines, and further developed by Dickinson and Jinks (1956) for heterozygous material, makes use of the array variances ($\bar{V}r$), and covariances ($\bar{W}r$) of arrays with the non-recurrent parent. The graphs are based on the joint analysis of the

FIG. 1.— W_r/V_r and W_r/W_r graphs for heavy seed set. Parental numbers as listed in table 1.

mean of reciprocals to minimise the effect of reciprocal differences. All graphs have been put on the same scale by multiplying Wr and Vr by $\frac{1}{V_p}$ (Allard, 1956), but not corrected for environmental factors (E_2). Only the graphs of F_1 data are presented, these are similar to those of the initial cross, but lacking any disturbance referable to the large (c) item.

The regression of Wr on Vr is non-significant (fig. 1), indicating either the absence of dominance, or considerable gene interaction. In the first case, the points should cluster at random around the position $Wr = 2Vr$ on the limiting parabola, this is not so, indicating that non-additive gene interaction is occurring. The distribution of the nine points follows no pattern but show considerable spurious overdominance effects (Jinks, 1955), which may be further inflated by environmental components (Jinks, personal communication).

The $W'r/Wr$ graph (fig. 1) can also be interpreted genetically (Hayman, 1954*b*, 1958), the theoretical regression of $+0.5$ is considerably less affected by genetic disturbances than the Wr/Vr graph, and virtually unaffected by the level of inbreeding. It may be used in the same way to detect the order of dominance in the parents. The $W'r/Wr$ graph differs from the Wr/Vr however in that it is affected by asymmetry of gene distribution, and indicates this whether the genes are correlated or not (Hayman, 1958). When there is asymmetry of gene distribution, parents with common genotypes will fall above the slope of $+0.50$, parents with different or relatively rare genotypes will fall below the line of $+0.50$. The graph (fig. 1) shows a wide scatter of points mainly above the theoretical line, with a considerable displacement of the *lusitanica* sample (3) well below the line.

(ii) Heavy seed germination

As seed of the northern populations required vernalisation (Borrill and Parker, in preparation), the germination data from the replicated experiment could not be used as a valid estimate of the genetic variation present; instead analysis was confined to a single vernalised replicate of 50 seeds of all parents and hybrids (table 4).

Hayman's analysis (table 5) shows that in the initial cross, only item (a) measuring additive genetic variation, and item (c) measuring consistent reciprocal differences, are significant when tested against (d). Item (c) is actually greater than (a), indicating the very strong effect of the cytoplasm in these crosses.

Germination of seed from the F_1 shows no consistent reciprocal effects, but both item (a) and (b) are significant. The Wr/Vr graph (fig. 2) again has no significant regression, in this case again indicating a considerable inflation of dominance effects caused by gene interaction and/or heterozygosity.

The $W'r/Wr$ graph (fig. 2) has a significant regression, (0.355 ± 0.097 ; $P = 0.01$) although there is a considerable scatter of points.

(iii) Fertility index

There is often a very high proportion of germinable underdeveloped seed in *Dactylis* chaff, which can alter the expression of fertility assessed as the number of live seedlings produced by a cross. Borrill (1961) used the

TABLE 4

Mean germination figures (converted to angles) of the initial crosses (upper line) and F₁ hybrids (lower line)

♂	♀ 1	2	3	4	5	6	7	8	9
1.	66.58 —	73.26 43.85	57.29 53.13	67.21 35.67	69.30 73.57	67.13 58.05	60.94 53.13	64.82 41.55	65.88 53.13
2.	47.35 66.42	63.43 —	62.87 43.85	77.89 50.77	73.26 88.19	59.08 63.43	56.42 48.45	65.88 69.73	70.54 66.42
3.	54.76 48.45	74.66 60.67	83.20 —	72.44 71.57	64.82 63.43	61.89 60.67	70.54 88.19	60.00 55.55	83.20 55.55
4.	33.71 33.21	52.89 53.13	54.03 77.08	66.89 —	88.19 53.13	60.73 53.13	65.35 43.85	64.75 66.42	48.73 60.67
5.	44.20 50.77	30.00 48.45	51.94 50.77	67.78 66.42	62.87 —	48.97 53.13	54.76 60.67	67.13 63.43	71.85 55.55
6.	49.78 43.85	49.78 63.43	69.30 63.43	68.28 58.05	59.08 48.45	62.17 —	62.87 69.73	68.11 73.57	62.87 46.15
7.	37.76 53.13	68.11 39.23	60.00 69.73	56.79 39.23	67.13 53.13	56.42 58.05	63.43 —	54.76 66.42	63.87 58.05
8.	60.00 41.55	60.00 60.67	70.54 66.42	77.08 60.67	64.82 55.55	57.29 41.55	62.87 73.57	75.82 —	73.26 66.42
9.	30.00 55.55	49.31 66.42	62.87 69.73	54.33 55.55	69.30 55.55	50.59 46.15	60.00 66.42	60.94 66.42	75.82 —

TABLE 5

Hayman's analysis of variance of unreplicated seed germination data for both initial cross, and F₁ hybrids

Item	D.F.	Initial cross		F ₁	
		M.S.	V.R.	M.S.	V.R.
<i>a</i>	8	174.91	2.86*	235.85	3.01*
<i>b</i>	36	79.43	1.30	158.27	2.02*
<i>c</i>	8	484.37	7.92***	92.33	1.18
<i>d</i>	28	61.14	—	78.31	—

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

fertility index to assess the potential contribution to the next generation, this is expressed by the following formulae:

(a)

$$\frac{\text{Total heavy seed} \times \% \text{Germination}}{100} = \text{Total seedlings (actual or potential)}$$

(b)

$$\frac{(\text{Total seedlings} + \text{Chaff seedlings})}{\text{Total No. of florets threshed}} \times 100 = \text{Fertility index}$$

FIG. 2.— $W'r/Vr$ and $W'r/W'r$ graphs for heavy seed germination.

This gives the number of live seedlings per 100 florets threshed. It is interesting to compare the analysis of this composite character with that of the major components. Initial cross and F_1 data are presented in table 6. Hayman's analysis (table 7) gives results closer to that of heavy seed set in

TABLE 6
Mean fertility index figures of the initial crosses (upper line) and F_1 hybrids (lower line)

♂	♀	1	2	3	4	5	6	7	8	9
1.		20.40 —	32.09 16.27	8.72 30.11	5.05 2.33	13.36 8.87	9.96 4.34	15.03 3.17	30.67 1.99	7.50 12.56
2.		15.41 50.95	20.22 —	18.80 11.00	22.75 8.13	7.38 14.02	9.24 4.30	10.82 3.71	32.81 22.59	18.73 5.50
3.		12.99 12.10	36.68 6.67	58.83 —	32.35 22.91	18.43 14.74	21.01 19.42	27.19 21.19	23.79 27.84	38.79 27.09
4.		7.48 2.75	44.95 4.41	34.51 18.34	21.18 —	33.02 11.96	20.88 2.01	19.68 4.30	29.11 3.16	21.43 2.13
5.		1.37 4.78	2.77 9.87	23.56 22.45	35.99 21.38	25.16 —	9.26 3.00	18.69 18.00	44.34 16.54	55.88 7.88
6.		3.76 4.95	18.71 0.99	45.67 21.78	27.59 2.13	12.05 2.10	20.95 —	13.91 5.17	25.29 16.57	28.73 1.41
7.		6.02 2.38	32.20 2.42	6.75 25.19	8.64 2.06	11.23 6.00	17.98 3.17	19.03 —	5.99 26.69	15.94 14.11
8.		12.38 2.46	39.16 17.71	23.29 23.71	26.73 2.01	17.13 7.01	12.00 4.12	12.27 41.82	38.13 —	34.35 19.59
9.		2.02 8.38	6.52 6.67	24.30 31.10	14.09 3.18	23.30 5.70	13.88 1.41	26.32 12.57	21.26 24.46	40.37 —

TABLE 7
Hayman's analysis of variance of unreplicated fertility index data for both initial cross, and F_1 hybrids

Item	D.F.	Initial cross		F_1	
		M.S.	V.R.	M.S.	V.R.
<i>a</i>	8	445.63	8.17***	508.75	16.11***
<i>b</i>	36	152.08	2.79**	178.59	5.66***
<i>c</i>	8	201.19	3.69**	47.98	1.52
<i>d</i>	28	54.52	—	31.57	—

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

both generations, the Wr/Vr graph (fig. 3) lacks the effects of considerable genetic disturbances expressed by the main components, presenting a pattern more amenable to interpretation. The regression of Wr on Vr is significant ($b = 1.002 \pm 0.152$; $P = 0.001$) and the line cuts the Wr axis negatively indicating the presence of some overdominance. There is a good positive correlation of parental values and the order of points along the regression line ($r = 0.850^{**}$), clearly indicating that dominance is directional for low fertility.

FIG. 3.— W_r/W_r and W_r/W_r graphs for fertility index.

The $W'r/Wr$ graph (fig. 3) shows the points at the lower end of the graph falling along the theoretical line of $+0.50$, the points at the upper (recessive) end of the graph, notably those representing subspecies samples of *lusitanica* (3) and *castellata* (9), are strongly displaced downwards and to the right, suggesting that the high fertility of these two isolated populations has come about by accumulation and selection of genes not present in the other population samples tested. This will be discussed further below.

(iv) Growth rate

The data for this character are presented in table 8, and the corresponding analysis of variance in table 9. Main effects are tested against the mean block interaction as the error variances are homogeneous ($\chi^2_{(5)} = 4.0$; $P = 0.5$). Significant additive (a) and dominance (b) variation occurs between the populations. All the components of dominance are significant, the largest is (b_1) indicating the occurrence of heterosis. There is also evidence of asymmetrical gene distribution (b_2) and inconsistent dominance interactions (b_3). Items (c) and (d) are both significant, the (c) effect being greater than (d) but not significantly so. Wearden's (1964) test also shows item (a) to be significantly greater than (c) and the (b) items significantly greater than (d) although (b_1) is the only component of (b) which is of any size.

The Wr/Vr graph (fig. 4) shows a non-significant regression, the parental points being widely scattered. The effect could be caused by non-allelic interaction and/or heterozygosity of the gene system, plus disturbances caused by consistent reciprocal differences.

The $W'r/Wr$ graph (fig. 4) is also disturbed by interaction effects and there is no significant regression. The points representing the population of *ashersoniana* (4) and *himalayensis* (6) are both displaced well below the theoretical line of $+0.5$. These are the most northerly subspecies, suggesting that there has been differential selection for genes reducing winter growth in these populations, this asymmetry being revealed by hybridisation.

(v) Stem internode elongation

In the vegetative phase *Dactylis* normally resembles the usual type of tussock grass, elongation of the stem internodes occurring only after inflorescence initiation at the shoot apex. In subspecies *smithii* (1), however, vegetative internode elongation is the usual condition, a long period of indeterminate growth resulting in a trailing habit. The subspecies is unique in the genus for expression of the character, which is also present to a small extent in the main shoot of seedlings of subspecies *santai* (8). The data are presented in table 10, all those with no elongation were given a figure of one for computational purposes.

Each main effect in the Haymans analysis has been tested against the individual block interactions, the error variances being heterogeneous ($\chi^2_{(5)} = 86.5$; $P = 0.001$) (table 11). There is significant additive genetic variation between the populations (a), together with significant dominance variation (b). All the components of dominance are significant, of these (b_1) is the largest, indicating directional dominance. Both items (c) and (d) are significant, however they are small compared to the other components of the analysis.

TABLE 8

Mean figures for growth rate, expressed as the number of days from germination to leaf six maturity

♂	♀	1	2	3	4	5	6	7	8	9
1.	71.00	59.67	65.00	63.00	62.67	62.67	73.00	67.00	66.00	
2.	67.33	62.67	63.33	69.00	57.33	59.33	69.00	60.00	66.33	
3.	70.67	65.33	71.00	63.00	62.33	62.00	76.00	69.67	68.00	
4.	64.33	63.67	63.67	78.33	58.67	74.00	67.33	65.00	62.67	
5.	66.00	70.00	64.67	58.00	61.33	60.00	70.67	61.33	61.33	
6.	61.00	63.00	64.00	68.00	62.33	59.00	68.33	63.67	64.00	
7.	73.67	67.00	72.33	64.00	60.67	66.00	80.76	72.33	72.00	
8.	67.00	57.00	62.67	61.00	63.33	63.00	71.33	71.67	68.33	
9.	66.33	65.33	67.67	63.33	60.67	63.33	77.67	67.67	70.33	

TABLE 9

Hayman's analysis of variance of growth rate data. Wearden's test is included as (c) and (d) significant

Item	D.F.	M.S.	V.R.	Wearden's test
<i>a</i>	8	360.55	46.95***	$\frac{a}{c}$ 8.65**
<i>b</i> ₁	1	419.45	54.62***	$\frac{b_1}{d}$ 20.01***
<i>b</i> ₂	8	61.00	7.94***	$\frac{b_2}{d}$ 2.91*
<i>b</i> ₃	27	46.70	6.08***	$\frac{b_3}{d}$ 2.23*
<i>b</i>	36	60.24	7.84***	$\frac{b}{d}$ 2.87**
<i>c</i>	8	41.70	5.43***	$\frac{c}{d}$ 1.99 N.S.
<i>d</i>	28	20.96	2.73***	—
<i>t</i>	80	—	—	—
<i>B</i>	2	16.31	2.12	—
<i>Ba</i>	16	6.70	—	—
<i>Bb</i> ₁	2	4.86	—	—
<i>Bb</i> ₂	16	12.20	—	—
<i>Bb</i> ₃	54	8.53	—	—
<i>Bb</i>	72	9.24	—	—
<i>Bc</i>	16	3.88	—	—
<i>Bd</i>	56	7.03	—	—
<i>Bt</i>	160	7.68	—	—
Total	242	—	—	—

* P < 0.05; ** P < 0.01; *** P < 0.001.

FIG. 4.— W_r/V_r and W_r^*/W_r graphs for seedling growth rate.

TABLE 10

Mean figures for stem internode elongation. Measured as the distance in cm. from ground level to the base of leaf nine

♂	♀	1	2	3	4	5	6	7	8	9
1.	13.72	13.11	5.14	2.90	3.44	3.50	6.72	14.27	7.81	
2.	5.83	1.00	1.00	1.00	1.00	1.00	1.00	1.35	1.00	
3.	4.51	1.00	1.00	1.00	1.00	1.00	1.00	2.14	1.00	
4.	2.01	1.00	1.00	1.00	1.00	1.00	1.00	1.14	1.00	
5.	5.61	1.00	1.00	1.00	1.00	1.00	1.00	1.43	1.00	
6.	2.60	1.00	1.00	1.00	1.00	1.00	1.00	1.50	1.00	
7.	11.36	1.00	1.00	1.00	1.00	1.00	1.00	2.81	1.00	
8.	14.51	1.35	1.33	1.07	1.28	1.46	3.22	10.89	1.00	
9.	4.22	1.00	1.00	1.00	1.00	1.00	1.00	3.21	1.00	

TABLE 11

Hayman's analysis of variance for stem internode elongation.
Wearden's test is included as (c) and (d) significant

Item	N.	M.S.	V.R.	Wearden's test
<i>a</i>	8	232.491	1498.38***	$\frac{a}{c}$ 84.66***
<i>b</i> ₁	1	27.135	5680.26***	$\frac{b_1}{d}$ 6.29*
<i>b</i> ₂	8	10.209	56.20***	$\frac{b_2}{d}$ 2.36*
<i>b</i> ₃	27	19.383	347.47***	$\frac{b_3}{d}$ 4.49***
<i>b</i>	36	17.559	213.05***	$\frac{b}{d}$ 4.07***
<i>c</i>	8	2.746	6.35***	$\frac{c}{d}$ — N.S.
<i>d</i>	28	4.315	12.52***	—
<i>t</i>	80	—	—	—
<i>B</i>	2	0.584	—	—
<i>Ba</i>	16	0.155	—	—
<i>Bb</i> ₁	2	0.005	—	—
<i>Bb</i> ₂	16	0.195	—	—
<i>Bb</i> ₃	54	0.052	—	—
<i>Bb</i>	72	0.082	—	—
<i>Bc</i>	16	0.432	—	—
<i>Bd</i>	56	0.344	—	—
Total	242	—	—	—

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

The Wr/Vr graph (fig. 5) shows a significant regression of Wr on Vr ($b = 0.829 \pm 0.086$; $P = 0.001$) which cuts the Wr axis positively. The position of the points so close to the limiting parabola indicates a low level of dominance for the gene systems concerned, however the nature and distribution of the character suggests that this effect is due either to correlated gene distribution, or a duplicate gene system, both resulting in the underestimation of the level of dominance (Hill, 1964; Mather, 1967). The correlation of the parental value with ($Wr + Vr$) is significant and positive ($r = 0.909^{***}$), indicating dominance for short internodes.

The $W'r/Wr$ graph (fig. 5) shows the points of subspecies *smithii* (1) and *santai* (8) falling below the theoretical line. The regression (0.416 ± 0.007 ; $P = 0.001$) deviates significantly from $+0.5$ ($t_{17} = 0.01$).

(vi) *Leaf six area*

The area of the sixth seedling leaf (table 12) was computed from length by median width, using a correction factor (Kemp, 1964). Hayman's analysis of variance is presented in table 13, Bartlett's test for heterogeneity of the six-error variances gives $\chi^2_{(5)} = 6.6$, so each mean effect is tested against the common error variance (Bt).

There is good evidence of additive genetic variation between the populations (item *a*), together with some dominance (item *b*), and the presence of consistent reciprocal effects (item *c*). All the components of dominance are significant, (b_1) being the largest, indicating the presence of heterosis and directional dominance, with some dominance variation due to gene asymmetry (b_2) and specific gene interaction (b_3). Wearden's (1964) test shows the (*c*) item to be significantly greater than (*d*) indicating the presence of real consistent reciprocal differences.

Genetic analysis by the Wr/Vr method (fig. 6) gives a significant regression ($b = 0.580 \pm 0.060$; $P = 0.001$) which is significantly different from the theoretical value of ≈ 0.9 ($t_{17} = 7.00^{***}$). The regression also cuts the Wr axis positively, the position of the lower points suggesting that these populations have a higher number of additive genes, or a more heterozygous gene complement for this character. There is a strong positive correlation of parental leaf area values and $Wr + Vr$ ($r = 0.813^{**}$) indicating that dominance is in the direction of low leaf area. The $W'r/Wr$ graph (fig. 6) also has a significant regression ($b = 0.661 \pm 0.101$; $P = 0.001$), the distribution of parental array points is virtually identical to that found on the Wr/Vr graph.

(vii) *Tiller number*

The number of tillers present at leaf six emergence varies considerably between parental populations (table 14). The results of Hayman's analysis of variance (table 15) shows the error variances to be heterogeneous ($\chi^2_{(5)} = 11.8$; $P = 0.05-0.02$) therefore each main effect is tested against its own block interaction.

There is good evidence of additive genetic variation between the populations (item *a*), and some dominance effects (*b*). Only (b_2) indicating the presence of gene asymmetry, and (b_3) indicating some form of dominance interaction are significant. However, none of the dominance components are much larger than the significant (*d*) item, which together with the significant (*c*) item indicate the presence of inconsistent and consistent

FIG. 5.— W_r/V_r and W_r/W_r graphs for seedling stem internode elongation.

TABLE 12

Mean figures for leaf six area in mm.²

♂	♀ 1	2	3	4	5	6	7	8	9
1.	742·01	750·60	1231·29	571·22	855·68	888·46	887·56	1150·41	1082·36
2.	689·25	424·43	643·07	239·01	517·78	472·39	489·96	592·03	823·75
3.	1204·64	579·95	1043·53	590·65	672·41	595·57	935·30	1113·91	1139·05
4.	486·96	282·52	666·82	130·69	237·08	193·68	341·96	357·93	435·73
5.	667·32	298·54	666·19	196·09	369·15	304·05	431·23	549·11	648·12
6.	776·50	496·91	666·29	202·60	365·52	295·76	523·20	640·54	518·81
7.	873·05	581·77	1032·07	440·80	574·58	554·08	639·16	725·49	724·15
8.	1046·88	649·41	1044·24	363·55	529·32	634·56	691·43	788·94	1024·14
9.	1071·65	622·10	1173·83	455·03	716·47	558·34	799·31	1022·68	713·49

TABLE 13

Hayman's analysis of variance for leaf-six area

Item	D.F.	M.S.	V.R.
<i>a</i>	8	1,954,410	360·72***
<i>b</i> ₁	1	189,731	35·02***
<i>b</i> ₂	8	68,254	12·60***
<i>b</i> ₃	27	36,003	6·64***
<i>b</i>	36	47,440	8·76***
<i>c</i>	8	24,748	4·57***
<i>d</i>	28	6,564	1·21 NS
Total	80	—	—
Block	2	79,534	14·68***
<i>Ba</i>	16	7,449	—
<i>Bb</i> ₁	2	6,943	—
<i>Bb</i> ₂	16	5,854	—
<i>Bb</i> ₃	54	5,929	—
<i>Bb</i>	72	5,941	—
<i>Bc</i>	16	6,686	—
<i>Bd</i>	56	3,803	—
<i>Bi</i>	160	5,418	—
Total	242	—	—

* $P < 0·05$; ** $P < 0·01$; *** $P < 0·001$.

FIG. 6.— W'/Vr and $W''r/Wr$ graphs for area of the sixth leaf on the main stem.

TABLE 14

Mean figures for tiller number at the sixth leaf stage

♂	♀	1	2	3	4	5	6	7	8	9
1.		0.11	0.25	1.56	1.30	1.11	1.06	1.44	1.17	2.53
2.		1.67	0.22	2.57	1.08	1.36	1.83	1.61	0.47	1.69
3.		2.14	2.42	3.56	2.64	3.44	3.50	3.22	2.67	2.92
4.		0.92	0.36	3.00	1.78	2.00	2.17	1.39	1.36	1.94
5.		0.75	1.36	3.19	2.25	2.67	3.39	1.72	1.58	1.94
6.		1.36	1.30	3.08	1.61	2.94	2.50	2.78	2.64	2.70
7.		1.50	1.67	3.03	1.94	2.64	2.67	3.11	2.19	2.21
8.		0.72	0.64	2.61	1.75	1.92	2.50	1.80	1.78	2.36
9.		2.30	1.33	3.28	2.47	2.81	3.06	2.70	2.50	2.69

TABLE 15

Hayman's analysis of variance for mean tiller number. Wearden's test included as (c) and (d) significant

Item	N.	M.S.	V.R.	Wearden's test
<i>a</i>	8	19.534	204.58***	$\frac{a}{c}$ 47.30***
<i>b</i> ₁	1	0.037	0.20	$\frac{b_1}{d}$ — N.S.
<i>b</i> ₂	8	0.365	16.00***	$\frac{b_2}{d}$ — N.S.
<i>b</i> ₃	27	0.562	4.23***	$\frac{b_3}{d}$ 2.71*
<i>b</i>	36	0.504	4.59***	$\frac{b}{d}$ 2.43**
<i>c</i>	8	0.413	8.06***	$\frac{c}{d}$ — N.S.
<i>d</i>	28	0.207	2.41**	—
Total	80	—	—	—
Block	2	0.809	—	—
<i>Ba</i>	16	0.095	—	—
<i>Bb</i> ₁	2	0.183	—	—
<i>Bb</i> ₂	16	0.023	—	—
<i>Bb</i> ₃	54	0.133	—	—
<i>Bb</i>	72	0.110	—	—
<i>Bc</i>	16	0.051	—	—
<i>Bd</i>	56	0.086	—	—
Total	242	—	—	—

* P < 0.05; ** P < 0.01; *** P < 0.001.

reciprocal effects. These facts and the large size of item (a), suggest that the genetic variation is largely additive in effect, this is confirmed by Wearden's (1964) test.

The genetical situation revealed by the Wr/Vr analysis (fig. 7) is rather different from that of leaf area. The scatter of points quite close on either side of the limiting point on the parabola ($Wr = 2Vr$) suggests a low level of dominance and/or a high level of heterozygosity. The regression of Wr on Vr is significant ($b = 1.001 \pm 0.074$; $P = 0.001$) and cuts the Wr axis positively. There is no correlation of parental values on $(Wr + Vr)$, indicating ambidirectional dominance.

The $W'r/Wr$ graph (fig. 7) shows a regression ($b = 0.521 \pm 0.004$; $P = 0.001$) not significantly different from $+0.5$ and quite free from any disturbance.

4. DISCUSSION

The fertility data present a considerably disturbed pattern of genetic variation, this is particularly so in germination. A similar though not so extreme pattern of inheritance is expressed in seed set, where the disposition of the population points on the Wr/Vr graph show considerable spurious overdominance. Only the complex character "fertility index" gives a relatively undisturbed Wr/Vr graph showing some overdominance, and directional dominance for low seedling production, obviously there is considerable disturbance of the relational balance of the fertility gene systems from the different subspecies populations. Presumably selection within these populations is directional towards high viability, although this does not appear to be maximal in more than two populations, those of *lusitanica* (3) and *castellata* (9) both of which are found in considerably disturbed habitats, here the selective advantage would tend to rise with increased viability. The $W'r/Wr$ graphs for germination and "fertility index" suggest that there is asymmetry of gene distribution involving these two populations.

The only comparable analysis of germination data, in perennial populations of *Lolium* (Hayward and Breese, 1966), show no consistent reciprocal effects and considerable dominance directional for high germination capacity. This presumably represents the difference between populations selected for asexual reproduction, and those subjected to selection for maximum seed production in strikingly different habitats.

Growth rate is considerably higher in the hybrids than in the parental subspecies samples, heterosis occurring frequently. Considering the fact that all the populations are outbreeding and reproduced by seed, this heterosis suggests that there may be a high proportion of sib-mating occurring, and despite some form of balancing selection in favour of heterozygotes, a consequent reduction of vigour below the maximum possible. This is made more likely by the fact that plants often occur in small, spatially isolated groups.

In their study of *Secale* species, Sun and Rees (1963) produced data for reduced growth rate of many hybrids in comparison with the parents. As some of the *Secale* species were inbreeders, and others outbreeders, this must be a functional difference in the level of genetic balance and differentiation between the two series, also differences in life cycle, and perhaps

FIG. 7.— Wr/Vr and Wr/Wr graphs for the number of tillers at the sixth leaf stage.

population size. In this case the *Dactylis* data follows the behaviour of hybrids between inbred lines (*vide* "April height" Bond, 1966).

Internode elongation, in showing a geographically orientated directional dominance of normal stem type, the most northerly populations expressing the highest number of dominant genes, indicates the strong selective disadvantage of this specialised character in any but the habitat in which it evolved. One interesting point is the low level of dominance mentioned earlier, although heterozygosity causes a reduction in the apparent level of dominance, and increases the suggestion of asymmetrical gene distribution, in this case the known distribution of the character implies that this apparently low dominance level is caused either by a system of associated genes or by duplicate gene interaction.

The dominance of low seedling leaf area is also closely related to climatic conditions experienced by the parental populations. Cooper (1964) has shown that seedling leaf expansion in both *Lolium* and *Dactylis* is highly correlated with the mean temperature of the coldest month. It is of interest that although a dominant gene system suppressing winter growth has evolved in these populations of the northern woodland subspecies, dominance is far from being complete, even though the character is of paramount survival value. Leaf area being correlated with leaf length ($r = 0.864^{***}$), comparison with the results of Hayward and Breese (1966) is possible. In north temperate pasture ecotypes of *Lolium perenne* there is dominance for short leaf length; however considerable genetic variation for the character exists between these already short leaved ecotypes, it is doubtful if any of them are homozygous for these differences. This also appears to apply to the north temperate woodland forms of *Dactylis*.

Tiller number also shows a low level of dominance, which is ambidirectional, suggesting stabilising selection within each parental population (Breese and Mather, 1960).

The results of the present study in subspecies reproducing by seed point to persistent and often large, consistent and inconsistent genetic effects in the seed and seedling characters measured; in only one case (leaf area), is there no gene/cytoplasm interaction. The considerable spurious overdominance expressed by the components of fertility, and viability (growth rate) works in opposite ways, the fertility genes balanced for (presumably) maximum production in the parental environment show hybrid disadvantage, the gene systems for vigour show hybrid advantage. This could be an unexpected consequence of the fertility "seive" through which these zygotes have passed, those surviving being formed by physiologically compatible gametes having complementary gene systems for vigour.

Additive effects are most important in the remaining characters, there being directional dominance for the two selectively important characters leaf area and internode length, and ambidirectional dominance for tiller number. These and following analyses should be a help towards gaining a better understanding of the nature of the subspecific differences in *Dactylis*.

5. SUMMARY

1. The genetic bases of differentiation in seed and seedling characters in one population of each of nine subspecies of diploid *Dactylis*, have been examined by means of the diallel cross analysis.

2. Analysis of the hybrids showed directional dominance for low fertility, and large additive components in the other characters with directional dominance in internode elongation and leaf area, and ambidirectional dominance in tiller number.

3. Both consistent and inconsistent reciprocal effects were present to a considerable degree in all the characters examined.

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