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TWO-LEVEL DIALLEL EXPERIMENTS INCLUDING PARENTS

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ABSTRACT

A two-level diallel mating design involving parental lines has been discussed. The analysis has been presented for randomised block design to control soil heterogeneity, if any. This design yields information about various types of combining abilities at population as well as individual levels and makes it possible to study the heterotic effects directly.

Key words: Average performance, combining abilities, inter- and intrapopulation hybridization, two-level diallel.

Tree improvement is one area in which genetic knowledge has played negligible role so far. It has been emphasized [1, 2] to carry out progeny tests for information on general and specific combining abilities (gca and sca) of the parent trees for seed orchards and to obtain reliable estimates of genetic parameters in order to decide the material and strategy for further breeding work. Among the many available mating designs, various forms of diallel crosses have been advocated [3–5] and actually used in forest tree breeding [6].

The above quoted work relates primarily to evaluation of performance of the clones or inbred lines on an intra- or intraspecies basis. However, as population, species and racial hybridization becomes more important [7] it becomes imperative to evaluate the performance of parents on an interpopulation or interspecies basis [8]. The work reported by [9, 10] was a step in this direction where crosses between two crops with and without parental lines were considered. A detailed study of two- level diallel cross experiment using information from intercrosses among m populations, each consisting of either n monoecious individuals or n moles and n females, has been reported in [11]. Such populations may represent species, races, clones, subpopulations or genetically meaningful collections of individuals. For example, five different species of *Pinus strobus; P. parniflora* Seeb. and Zucc., *P. peuce* Griseb., *P. griffithii* McClel., *P. monticola* D. Don, and *P. strobus* L., have produced successful interspecific crosses [8, 12].

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The present work is an extension of [11] so as to include parental lines and conduct of the experiment in randomised blocks to control soil heterogeneity, if any. The inclusion of parents is desirable because it provides a means of estimating hybrid vigour directly from the specific combining effects. We shall, however, presently deal with the methodological aspects of the experiment, its application in wheat is proposed to be given in a subsequent paper. A computer software for such experiments has also been prepared [13].

MODEL AND NOTATIONS

For conformity of notations with those in [11], we consider m populations (species or groups) P_i ($i = 1, 2, \ldots, m$), each having n individuals (clones or lines) I_{ip} ($p = 1, 2, \ldots, n$). The types of matings to be considered in this paper are diagrammed in Fig. 1 for $m = 3$ and $n = 4$.

Individual	Populations											
	P_1				P_2				P_3			
	I_{11}	I_{12}	I_{13}	I_{14}	I_{21}	I_{22}	I_{23}	I_{24}	I_{31}	I_{32}	I_{33}	I_{34}
I_{11}	*				*	*	*	*	*	*	*	*
I_{12}		*			*	*	*	*	*	*	*	*
I_{13}			*		*	*	*	*	*	*	*	*
I_{14}				*	*	*	*	*	*	*	*	*
I_{21}					*				*	*	*	*
I_{22}						*			*	*	*	*
I_{23}							*		*	*	*	*
I_{24}								*	*	*	*	*
I_{31}									*			
I_{32}										*		
I_{33}											*	
I_{34}												*

Fig. 1. Crossing pattern among 3 populations and 4 individuals in each population.

Let $Y_{(ip)\,(jq)z}$ be the observation on the progeny between a cross of pth individual belonging to ith population and qth individual belonging to jth population (i.e. $I_{ip} \times I_{jq}$) in zth replicate ($z = 1, 2, \ldots, r$). In case of more progeny per cross, this can be taken as mean over the progenies. An appropriate linear model for this design is

$$Y_{(ip)(jq)z} = M + Bz + G_i + G_j + S_{ij} + g_{ip} + g_{jq} + S_{(ip)(jq)} + e_{(ip)(jq)z}$$

where M—general mean, B_z—zth block effect, G_i—general combining ability (gca) effect of P_i, S_{ij}—specific combining ability (sca) effect of the cross $P_i \times P_j$, g_{ip}—gca of I_{ip}, $s_{(ip)(jq)}$—sca of the cross $I_{ip} \times I_{jq}$, and $e_{(ip)(jq)z}$—a random competent assumed to be distributed normally and independently with mean zero and constant variance $\sigma^2 e$.

The following notations are used to simplify the computations:

$$Y(..)(..)z = \sum_{i,j} \sum_{p,q} Y_{(ip)(jq)z} + \sum_{i,p} Y(ip)(ip)z$$

$$Y(ip)(jq). = \sum_{z} Y(ip)(jq)z$$

$$Y(i.)(j.). = \sum_{p,q} Y(ip)(jq)., (i \neq j)$$

$$Y(i.). = \sum_{p} Y(ip)(ip).$$

$$Y(ip)(..). = \sum_{j,q} Y(ip)(jq). + 2\, Y(ip)(ip).$$

$$Y(i.)(..). = \sum_{p}^{j \neq i} Y(ip)(..).$$

$$Y(..)(..). = \sum_{z} Y(..)(..)z$$

Throughout the text the suffixes will run as: i, j = 1, 2, ..., m; p, q = 1, 2, ..., n; and z=1, 2, ..., n.

ANALYSIS TECHNIQUES

Depending on the type of inference, we discuss here two models:

(i) The fixed effects model: B_z, M, G_i, S_{ij}, g_{ip} and $s_{(ip)(jq)}$ are fixed effects with

$$\sum_{z} B_z = 0 = \sum_{i} G_i ; \sum_{p} g_{ip} = 0 \text{ for every i}$$

$$n \sum_{\substack{j \\ j \neq i}} S_{ij} + 2\, S_{ii} = 0, \text{ for every } i \; (S_{ij} + S_{ji})$$

$$\sum_{p} s_{(ip)(ip)} = 0, \text{ for every } i$$

$$\sum_{p,q} s_{(ip)(jq)} = 0, \text{ for every } (i, j)$$

$$\sum_{\substack{j,q \\ j \neq q}} s_{(ip)(jq)} + 2\, s_{(ip)(ip)} = 0, \text{ for every } (i, p)$$

$$(s_{(ip)(jq)} = s_{(jq)(ip)})$$

(ii) The random effects model: B_z, G_i, S_{ij}, g_{ip} and $s_{(ip)(jq)}$ are random variables, independently distributed with means zero and variances σ^2B, σ^2G, σ^2S, σ^2g and σ^2s, respectively.

In (i), the inference is made only with respect to the populations and individuals actually included in the experiment, whereas in (ii) inference is made with regard to a larger collection of populations and individuals of which those included in the experiment represent a random sample.

The least square estimators of different parameters and their variances are given in Table 1. These expressions are needed when one compares the effects against their hypothesized values, such as a zero value of gca or sca. It may be emphasized that for testing against a zero value the proper hypothesis should be a one-tailed test, namely,

$$HO: g = 0, Hi: g > 0 \text{ if } \hat{g} > 0, \text{ or}$$

$$HO: g = 0, Hi: g < 0 \text{ if } \hat{g} < 0$$

Perhaps more important aspect of the experiment lies in comparing different effects. For this purpose different comparisons and their variances are listed in Table 2. A word of caution: the Student's least significant difference (LSD) method should be used for paired comparisons only if the number of entries to be compared does not exceed 5 or 6, otherwise one should go for the multiple range test [14, 15], since in former case the probability of rejection exceeds far beyond the limit fixed as size of the test [16, 17]. Analysis of variance (ANOVA) and expectations under the fixed and random effects models are given in Tables 3 and 4, respectively, for testing the hypotheses of interest.

Table 1. Least squares estimators and their variances

Estimator	Variance/σ_e^2
$\hat{M} = 2\,Y(..)(..).\,/a^*mnr$	—
$\hat{B}z = 2\,Y(..)(..)z/amn - 2\,Y(..)(..)./amnr$	—
$\hat{G}_i = \frac{Y(i.)(..).}{b^*nr} - \frac{2Y(..)(..).}{bmnr}$	$\frac{m-1}{bmnr}$
$\hat{S}_{ij} = \frac{Y(i.)(j.)}{n^2r} - \frac{Y(i.)(..) + Y(j.)(..)}{bnr} + \frac{2\,Y(..)(..).}{abr}$	$\frac{n(m-2)\ (m-3)\ n + b\ + 8}{abn^2\, r}$
$\hat{S}_{ii} = \frac{Y(i.)(i.).}{nr} - \frac{2Y(i.)(..).}{bnr} + \frac{2Y(..)(..).}{bar}$	$\frac{(m-1)\ (m-2)\ n + 2}{abr}$
$\hat{g}_{ip} = \frac{Y(ip)(..).}{cr^*} - \frac{Y(i.)(..).}{cnr}$	$\frac{n-1}{cnr}$
$\hat{s}_{(ip)(jq)} = \frac{Y(ip)(jq).}{r} - \frac{Y(ip)(..).+Y(jq)(..).}{cr} + \frac{Y(i.)(..).+Y(j.)(..).}{cnr} - \frac{Y(i.)(j.).}{n^2r}$	$\frac{c(n^2-1) - 2n\ (n+1)}{cn^2\, r}$
$s(ip)(ip) = \frac{Y(ip)(ip).}{r} - \frac{2Y(ip)(..).}{cr} + \frac{2Y(i.)(..).}{cnr} - \frac{Y(i.)(i.).}{nr}$	$\frac{(m-1)(n-1)}{cr}$

*a—(m–1) n+2, b—(m–2) n+4, c—(m–1) n+4.

COMBINING ABILITY INFORMATION FROM THE EXPERIMENTS

Estimators given in Table 1 or comparisons between two estimators (Table 2) can be tested against their hypothesized values using the given expressions for variances under the fixed effects model. The unknown $\sigma^2 e$ will be substituted for its estimate. Another important comparisons may be of the following types:

i) Average performance of Pi vs. average performance of Pj

ii) Average performance of hybrid population ($P_i \times P_j$) vs. average performance of hybrid population ($P_i' \times P_j'$)

Table 2. Variances of comparisons between combining abilities in σ_e^2 units

Comparison	Variance
$\hat{G}_i - \hat{G}_i{}'$*	2 /bnr**
$\hat{S}_{ij} - \hat{S}_{i}{}'{}_j$	$2\{(m-3)n+4\}/bn^2r$
$\hat{S}_{ij} - \hat{S}_{i}{}'{}_{j}{}'$	$2\{(m-4)n+4\}/bn^2r$
$\hat{S}_{ii} - \hat{S}_{i}{}'{}_{i}{}'$	$2(m-2)/br$
$\hat{S}_{ii} - \hat{S}_{ij}$	$\{b(n+1)-2n\}/bn^2r$
$\hat{S}_{ii} - \hat{S}_{i}{}'{}_j$	$\{b(n+1)-6n\}/bn^2r$
$\hat{g}_{ip} - \hat{g}_{ip}{}'$	$2/cr$
$\hat{g}_{ip} - \hat{g}_{i}{}'{}_{p}{}'$	$2(n-1)/cnr$
$\hat{s}(ip)(jq) - \hat{s}(ip)(j'q')$	$2\{(m-1)n+3\}/cr$
$\hat{s}(ip)(jq) - \hat{s}(ip')(jq')$	$2a/cr$
$\hat{s}(ip)(jq) - \hat{s}(ip)(j'q')$	$2(n-1)\{c(n+1)-n\}/cn^2r$
$\hat{s}(ip)(jq) - \hat{s}(ip')(j'q')$	$2\{c(n^2-1)-n(2n-1)\}/cn^2r$
$\hat{s}(ip)(jq) - \hat{s}(i'p')(j'q')$	$2\{c(n^2-1)-2n(n-1)\}/cn^2r$
$\hat{s}(ip)(ip) - \hat{s}(ip)(jq)$	$\{c(2n^2-n-1)-2n(n-2)\}/cn^2r$
$\hat{s}(ip)(ip) - \hat{s}(ip')(jq)$	$\{c(2n^2-n-1)-2n(3n-1)\}/cn^2r$
$\hat{s}(ip)(ip) - \hat{s}(i'p')(j'q')$	$\{c(2n^2-n+1)-6n(n-1)\}/cn^2r$
$\hat{s}(ip)(ip) - \hat{s}(ip')(ip')$	$2(m-1)n/cr$
$\hat{s}(ip)(ip) - \hat{s}(i'p')(i'p')$	$2(m-1)(n-1)/cr$

*It has been assumed that $i' \neq i$, $j' \neq j$ etc.

**a, b and c as in Table 1.

iii) Average performance of I_{ip} vs. average performance of $I_{i'p'}$)

iv) Average performance of hybrid ($I_{ip} \times I_{jq}$) vs. average performance of hybrid ($I_{i'p'} \times I_{j'q'}$)

These comparisons along with their estimators and variances are given in Table 5. As for the case of random effects model, the estimates of the variance components can be had, as usual, from Table 4. However, making a correspondence between covariances of individuals at different population levels and variance components is quite involved and in under study [6].

Table 3. Analysis of variance

Source	Degree of freedom	Sum of squares	Mean squares
B	r–1	$\frac{2}{amn}\sum_{z} Y^2(..)(..). - \frac{2A^*}{amnr}$	MS(B)
G	m–1	$\frac{1}{bnr}\sum_{i} Y^2(i.)(..). - \frac{4A}{bmnr}$	MS(G)
S	$\frac{m(m-1)}{2}$	$\frac{1}{n^2 r}\sum_{\substack{i,j\\i<j}} Y^2(i.)(j.). + \frac{1}{nr}\sum_{i} Y^2(i.)(i.).$	MS(S)
		$- \frac{1}{bnr}\sum_{i} Y^2(i.)(..). + \frac{2A}{abr}$	
g	m(n–1)	$\frac{1}{cr}\sum_{i,p} Y^2(ip)(..). - \frac{1}{cnr}\sum_{i} Y^2(i.)(..).$	MS(g)
s	$\frac{m(m-1)}{2}(n^2-1)$	$\frac{1}{r}\sum_{\substack{i,j\\i<j}}\sum_{p,q} Y^2(ip)(jq). + \frac{1}{r}\sum_{i,p} Y^2(ip)(ip).$	MS(s)
		$- \frac{1}{cr}\sum_{i,p} Y^2(ip)(..). + \frac{1}{cnr}\sum_{i} Y^2(i.)(..).$	
		$- \frac{1}{n^2 r}\sum_{\substack{i,j\\i<j}} Y^2(i.)(j.). \quad \frac{1}{nr}\sum_{i} Y^2(i.)(i.).$	
e	$\{m\frac{(m-1)}{2}n^2$ + mn–1)} (r–1)	By subtraction	MS(s)
Total	$\frac{m(m-1)}{2}n^2r$ + mnr — 1	$\sum_{\substack{i,j\\i<j}}\sum_{p,q,z} Y^2_{(ip)(jq)^z} + \sum_{i,p,z} Y^2_{(ip)(ip)z}$ i–j $- \frac{2A}{amnr}$	

$^*A = Y^2(..)(..).$; a, b, c as in Table 1.

HETEROSIS STUDIES

In view of the information available on parents, it is possible to study heterotic effects directly. These can be computed as percentage increase or decrease in mean values of the hybrids over their midparents, better parent or best parent as the investigator wishes, and can be tested with appropriate standard errors. A χ^2 contingency is suggested to test the

Table 4. Expected mean squares

Mean squares	Model I	Model II
MS (B)	$\sigma_e^2 + \frac{a^*}{2(r-1)} mn \sum_Z B_z^2$	$\sigma_e^2 + \frac{a}{2} mn\, \sigma_B^2$
MS (G)	$\sigma_e^2 + \frac{b}{(m-1)} nr \sum_i G_i^2$	$\sigma_e^2 + r\, \sigma_s^2 + b\, r\, \sigma_g^2 +$
		$\frac{(m-2)\, n^2 + 4}{b}\, nr\, \sigma_S^2 + bnr\, \sigma G^2$
MS (S)	$\sigma_e^2 + \frac{2nr}{m(m-1)} \left\{ n \sum_{i<j} s_{ij}^2 + \sum_i S_{ii}^2 \right\}$	$\sigma_e^2 + r\, \sigma_s^2 + \frac{(m-2)[(m-3)n^2 + 8n]+12}{a-b}\, n^2 r\, \sigma_S^2$
MS (g)	$\sigma_e^2 + \frac{cr}{m(n-1)} \sum_{i,\,p} g_{ip}^2$	$\sigma_e^2 + r\sigma_s^2 + c\, r\sigma_g^2$
MS(s)	$\sigma_e^2 + \frac{2r}{m(m-1)\,(n^2-1)}\, X$	$\sigma_e^2 + r\, \sigma_s^2$
	$\left\{ \sum_{i<j} \sum_{p,\,q} s^2_{(ip)\,(jq)} + \sum_{i,\,p} s^2_{(ip)\,(ip)} \right\}$	
MS(e)	σ_e^2	σ_e^2

*a, b and c as in Table 1.

Table 5. Comparisons of average performance

Type of comparison	Estimator	Variance
$G_i - G_i'$	$\{Y_{(i.)\,(..).} - Y(i)\ (..).\}/bnr^*$	$2\sigma_e^2/bnr$
$(G_i + G_j + S_{ij})$ $- G_i' + G_j' + S_{i'j'})$	$\{Y_{(i.)\,(j.).} - Y_{(i.')\,(j.')}.\}/n^2 r$	$2\ \sigma_e^2/n^2\, r$
$(G_i + g_{ip}) - (G_i' + g_{i'p'})$	$\{Y_{(ip)\,(..).} - Y_{(i'p')\,(..)}.\} / (a+2)\, r$ $+ \{Y_{(i.)\,(..).} - Y_{(i.')\,(..)}.\} / (a+2)\, br$	$2\, \{(a+3)\, b+n\}\, \sigma_e^2\ /\ (a+2)\, br$
$\{G_i + G_j + S_{ij} + g_{ip}$ $+ g_{jq} + s_{(ip)\,(jq)}\} -$ $\{Gi' + Gj' + Sij' + gi'p'$ $+ gi'q' + s(i'p')\,(j'q')\}$	$\{Y_{(ip)\,(jq)}. - Y_{(i'p')\,(j'q')}.\} / r$	$2\ \sigma_e^2/r$

*a, b as in Table 1.

hypothesis whether high/low heterotic cross combinations are associated with the parents of high/low gca effects or high/low sca effects [16]. This aspect will be discussed in detail in a subsequent article with data on wheat.

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