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MISSING PLOT TECHNIQUE FOR DIALLEL ANALYSIS

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ABSTRACT

The method of analysis of diallel crosses when one or two observations are missing has been developed for Griffing Method I, II and III. The formulae for obtaining estimates of various parameters involving the missing line or otherwise along with their estimates of variances are given. The ANOVA tables for combining ability analysis for all the three methods are also presented. It has been observed that extension of the technique for more than two missing observations is quite simple and straightforward.

Key words: Diallel crosses, gca, sca, reciprocal differences, Bartlett missing plot technique.

Diallel analysis is very commonly used in plant and animal breeding for identifying the best selection potential of the crosses in an early generation. Griffing [1) proposed four methods of analysis depending on the inclusion of parents and/or reciprocals along with F1S. The breeders are unable to perform combining ability analysis of diallel crosses when some observations are missing which sometimes happens in field experiments due to several natural hazards.

The use of missing plot technique has been quite common in environmental designs but its use in mating designs, like diallel crosses has been very limited. Kaushik & Puri [2, 3] suggested a missing plot technique for diallel crosses based on Topham model (4). Manocha (5] proposed a missing plot technique in diallel crosses for Method-IV of Griffing (1]. In the present paper, the methods of analysis of remaining three methods, i.e. Methods I, II and III ofGriffing [l], when one or two observations are missing, based on the Bartlett (6] missing plot technique have been presented. In case of one missing observation it may pertain to a cross or to a parent in Method I and Method II, while in Method III it will pertain to a cross only. These cases will be discussed together.

MODEL AND ANALYSIS WHEN ONE OBSERVATION IS MISSING

Consider n possible crosses of v inbred lines laid out in randomized block design with r replications.

70 L. S. *Kaushik et al.* [Vol. 57, No.1

Let the observation yi'j'k' in a cross (i' x j') involving i'-th female mated to j'-th male in k'-th block be missing. If $i' = j'$, the missing observation pertains to the i' -th parent. The model to be considered for combining ability analysis in Method I will be as under:

$$
y_{ijk} = \mu + g_i + g_j + s_{ij} + r_{ij} + \beta(x_{ijk} - \bar{x}) + e_{ijk}
$$

(1)
(i, j = 1, 2, 3 ... v; k = 1, 2 ... r)

where y_{ijk} is the observation on $(i \times j)$ th cross, i.e. i-th female mated to j-th male in k-th block; μ is the average effect; g_i (g_i) is the gca effect of i-th (j-th) line; sij is the sca effect of (i x j)th cross; r_{ij} is the reciprocal effect; b_k is the k-th block effect; x_{ijk} is the corresponding observation of concomitant variable; β is the regression coefficient of y on x; and e_{ijk} is the random error associated with the observation y_{ijk} assumed to be normally and independently distributed with mean 0 and constant variance σ^2 , x_{ijk} will have the value 1 if only one observation is missing and 0 otherwise and \bar{x} is the overall mean of concomitant variable. The corresponding value of y for the missing case will be taken as zero.

Using the notation of Griffing [1], the estimates of parameters are obtained by the method of least squares as given below:

Method I

$$
\hat{\mu} = y \dots / rv^2
$$
\n
$$
\hat{g}_i = (y_{i.} + y_{.i})/2vr - y \dots / rv^2 - a_i \, (8)\hat{\beta}/rv^2
$$
\n
$$
\hat{s}_{ij} = (y_{ij.} + y_{ji.})/2r - (y_{i..} + y_{.i.} + y_{.j.} + y_{.j.})/2vr + y \dots / rv^2 - a_{ij} \, (s) \, \hat{\beta}/rv^2
$$
\n
$$
\hat{s}_{ii} = y_{ii.}/r - (y_{i..} + y_{.i.})/vr + y \dots / rv^2 - a_{ii} \, (s) \, \hat{\beta}/rv^2
$$
\n
$$
\hat{r}_{ij} = (y_{ij.} - y_{ji.})/2r - a_{ij} \, (r) \, \hat{\beta}/2r
$$
\nMethod II\n
$$
\hat{\mu} = 2y \dots / rv \, (v + 1)
$$
\n
$$
\hat{g}_i = [v(y_{i..} + y_{ii.}) - 2y \dots - a_i \, (8)\hat{\beta}]/rv(v + 2)
$$
\n
$$
\hat{s}_{ij} = [(v + 1) (v + 2)y_{ij.} - (v + 1) (y_{i..} + y_{ji.} + y_{j..} + y_{jj.}) + 2y \dots - a_{ij} \, (s)\hat{\beta}]/r(v + 1) \, (v + 2)
$$
\n
$$
(i \neq j)
$$

 $S_{ii} = [(v+1) (v+2)y_{ii} - 2(v+1) (y_{i..} + y_{ii.}) + 2y ... - a_{ii} (s)\hat{\beta}]/r(v+1) (v+2)$ (3)

(2)

February, 1997)

Missing Plot Technique in Diallel Analysis 71

Method III

$$
\hat{\mu} = y.../rv (v-1)
$$
\n
$$
\hat{g}_i = [v(y_{i..} + y_{i.}) - 2y... - a_i \otimes \hat{f}]/2rv (v-2)
$$
\n
$$
\hat{s}_{ij} = (y_{ij.} + y_{ji})/2r - (y_{i..} + y_{i..} + y_{j..} + y_{j.})/2r(v-2) + y... /r(v-1) (v-2) - a_{ij} \otimes \hat{f} / 2r(v-1) (v-2) \qquad (i \neq j)
$$
\n
$$
\hat{r}_{ij} = (y_{ij.} - y_{ji.})/2r - a_{ij} \otimes \hat{f} / 2r \qquad (4)
$$

In the above formulae $a_i^{(g)}$, $a_{ii}^{(s)}$, $a_{ii}^{(s)}$ and $a_{ii}^{(r)}$ are the constants which will have the values as specified in Table 1. The estimate of regression coefficient is obtained as under:

$$
\beta = E_{xy}/E_{xx} \tag{5}
$$

where $E_{xy} = (y \dots -ry \dots k'-ny_{i'j'})/nr$. and $E_{xx} = (r-1) (n-1)/nr$. The estimates of block effects are not given as they are of no interest to a geneticist or breeder.

From the above estimates, one has to pick up the corresponding applicable estimates depending on the method used and whether the observation on a cross or a parent is missing. Now let us consider the case when the observation on $(i' x j')$ th cross or a parent in Method I is missing. Here $a_i^{(g)}$ will have only two values, (v-2)/2 and -1, depending on the involvement or no involvement of i-th line in missing cross. Consequently, the estimate of gi will be:

$$
\hat{g}_i = (y_{i..} + y_{.i.})/2vr - y_{.} \dots /rv^2 - (v-2)\hat{\beta}/2rv^2 \quad \text{if } i = i'
$$
\n
$$
\hat{g}_i = (y_{i..} + y_{.i.})/2vr - y_{.} \dots /rv^2 + \hat{\beta}/rv^2 \quad \text{if } i \neq i'
$$

For estimating s_{ij} , a_{ij} (s) will have all values mentioned in the table except $-(v-1)$ depending on whether both i-th lines or only one or none of them is involved in missing observation. Similarly a_{ii} ^(r) will have three values and, consequently, there will be three possible estimates of r_{ii} .

Now let us consider the case when an observation on i'-th parent in k'-th block is missing. For estimating g_i , a_i $^{(g)}$ will have values v–1 and 1; for estimating s_{ij} , a_{ij} $^{(s)}$ will have the values $-(v-1)$ and 1; for estimating s_{ii} , a_{ii} ^(s) will have the values $(v-1)^2$ and 1; and for estimating r_{ij} , a_{ij} ^(r) will have only zero value. Similarly, the estimates of parameters are obtained in the remaining two methods also. For instance, in Method II, in case of missing observation on a cross, there will be two estimates each of g_i and s_{ij} depending upon whether

the i-th line is involved or not in a missing cross; and three estimates of s_{ij} when both of the parents are involved in the missing cross. Similarly, if an observation on a parent is missing, there will be two estimates each of g_i , s_{ij} and s_{ii} . Likewise in Method III, there will be two possible estimates of g_i and three of s_{ij} as well as r_{ij} .

The sums of squares (SS) obtained by usual method of fitting constants given by Coons[7], are presented in Tables 2, 3 and 4 for Method I, Method II and Method III, respectively. The notations in the tables used are as under.

Parameter	Method I	Method II	Method III	Explanation
a_i ^(g)	$(v-1)$	$2(v-1)$	N.A.	If i-th line is involved in the missing observation on a parent $(i = i' = j')$
	$(v-2)/2$	$(v-2)$	$(v-2)$	If i-th line is involved in the missing observation on a cross $(i = i' or j', i' \neq i')$
	-1	-2	-2	Otherwise
a_{ij} ^(s)	$(v^2-2v+2)/2$	(v^2+v+2)	$(v-2)(v-3)$	If both i-th and j-th lines are involved in the missing observation $(i = i', j = j'$ or $i = j', j = i'$ $i' \neq j'$
	$-(v-2)/2$	$-(v-1)$	$-(v-3)$	If either i-th or j-th line is involved in the missing observation (i or $j = i'$ or j') or $(i' \neq j')$
	$-(v-1)$	$-2v$	N.A.	If observation on i-th or j-th parent is missing $(i = i' = j' \text{ or } j = i = j')$
	-1	$\overline{2}$	$\overline{2}$	Otherwise
a_{ii} ^(s)	$(v-1)^2$	$v(v-1)$	N.A.	If missing observation pertains to i-th parent $(i = i' = j')$
	$-(v-1)$	N.A. $-2v$	If missing observation involves i-th line as male or female parent $(i = i' or j', i' \neq j')$	
	1	$\overline{2}$	N.A.	Otherwise
a_{ii} ^(r)	1	N.A.	1	If observation on (i x j)th cross is missing (i = i', j = j')
	-1	N.A.	-1	If observation on $(i \times i)$ cross is missing
	0	N.A.	Ω	Otherwise

Table 1. Values of a_i (g), a_{ij} (s), a_{ii} (s) and a_{ij} (r) in three different diallel methods

N.A. - Not applicable.

Method I

$$
R_{yy} = \Sigma y^2 ... k/v^2 - y^2 ... / rv^2,
$$

\n
$$
C_{yy} = \Sigma y^2_{ij.} / r - y^2 ... / rv^2
$$

\n
$$
G_{yy} = \Sigma (y_{ij.} + y_{.i.})^2 / 2vr - 2y^2 ... / rv^2,
$$

\n
$$
r_{yy} = \Sigma (y_{ij.} - y_{ji.})^2 / 2r
$$

\n
$$
S_{yy} = \Sigma (y_{ij.} + y_{ji.})^2 / 2r + \Sigma y_{ii.}^2 / r - \Sigma (y_{i.} + y_{.i.})^2 / 2vr + y^2 ... / rv^2
$$

\n
$$
E_{yy} = \Sigma y^2_{ijk} - \Sigma y^2_{ij.} / r - \Sigma y^2_{..k} / v^2 + y^2 ... / rv^2
$$
\n(6)

February, 1997J *Missing Plot Technique in Dialle! Analysis* 73

Source	d.f.	Adjusted sum of squares
Replications	r-1	R_{vv} + βE_{xy} – y^2 is in /r(r–1)
Crosses	v^2-1	C_{vv} + β Exy – y^2 . k'/v ² (v ² –1)
Gca ⁺	$v-1$	G_{yy} + βE_{xy} - $[v(y_{i'} + y_{j'} + y_{j'} + y_{j'}) - 2ry \cdot k' - 2v^2y_{i'j'} - 2y \cdot j^2 / 4v^2r[(v-2) + (r-1)(v^2-1)]$
Sca [*]	$v(v-1)/2$	S_{yy} + $\hat{\beta}E_{xy}$ - $[v^2(y_{ij'}/-y_{ji'}) - v(y_{i'} + y_{i'} + y_{j'} + y_{j'})$ -2ry κ + $4y$ κ + $\frac{1}{2}/2v^2r [(v^2-2v+2) + 2(r-1)(v^2-1)]$
Reciprocals	$v(v-1)/2$	r_{vv} + βE_{xv} + $[2y/2ryk'-v^2(y_{i'j'}-y_{i'j'})]^2/2v^2r(rv^2-r+1)$
Error	$(r-1)(v^2-1)-1$	E_{vv} – βE_{xy}

Table 2. ANOVA table for Method I when observation on a cross is missing

"When observation on a parent is missing.

¹

 \int 2 2 \int 2 \int 2 \int (y_y + \int E_{xy} - [v(yi_{'..} + y_{'i'}) - ry \int_{x} _{k'} - v²y_{i'j'}.-2y ...¹² /v²r[2(v-1) + (r-1) (v²-1)] Sca SS = S_{yy} + β E_{xy} - [2y ... – v (y_{i'..} + y_{ii'}) – ry ..k']²/v²r[(rv² – 2v – r + 2) Reciprocal $SS = r_{yy}$

Table 3. ANOVA table for Method II when observation on a cross is missing

Source	d.f.	Adjusted sum of squares
Replications	r-1	R_{vv} + βE_{xy} – y^2 i' j'. /r(r–1)
Crosses	$v(v+1)/2-1$	C_{vv} + β Exy – 4y ² . k'/v(v+1) (v ² + v – 2)
Cca	$v-1$	G_{yy} + βE_{xy} – [v(v+1) (y _i + y _{ii} + y _j + y _j + y _j + v(v+1) (v+2)y _{ij} – 2(v+2)r y. .k' - 2vy] ² /vr(v+1) (v+2) [v ³ (r-1) + v ² (3r-1)-2v-4r]
Sca ⁻	$v(v-1)/2$	S_{yy} + βE_{xy} – $[4(v+1)y - v(v+1)(y_{i'_{-}} + y_{i'_{+}} + y_{j'_{-}} + y_{i'})]$ - $2r(v+2)yk'$ $i^2/vr(v+1)(v+2)$ $[rv^3 + v^2(3r-2) + 2v - 4(r-1)]$
Error	$[v(v+1)/2-1][r-1]-1$ $E_{vv} - \beta E_{xv}$	

'When observation on a parenl is missing.

Gca SS = G_{yy} + βE_{xy} - $\frac{1}{2}$ v(v+1) (y_j: + y_{i'i};) - v(v+1) (v+2) y_{i'i}: - 2(v+2) ry_{1k}'
- 2vy ...]²/vr(v+1) (v+2) {4(v²-1) + (r-1) (v+2) (v² + v-2)]

Sca SS = S_{yy} + βE_{xy} - [4(v+1)y . . . - 2v(v+1) (y_{i'.}, + y_{i'i'}) - 2r(v+2)y_{. k'}]²/vr(v+1) (v+2) [rv³ + v²(3r-4)-4(r-1)]

÷

Table 4. ANOVA table for Method III when observation on a cross is missins

Source	d.f.	Adjusted sum of squares
Replications	r-1	R_{vv} + βE_{xy} – y^2 $y y'$ /r(r–1)
Crosses	$v^2 - v - 1$	C_{vv} + β Exy – y ² . k'/v(v–1) (v ² –v–1)
Gca	$v-1$	G_{yy} + $\hat{\beta}E_{xy}$ - [v(v-1) (y _{i',} + y _{i',} + y _{i',} + y _i ', - 2v(v-1) (v-2)y _{i'j',} - 2r(v-2)y _{ik'} - y v] ² /4(v-2) ² [(v-1) + (r-1) (v ² -v-1)]
Sca	$v(v-3)/2$	S_{yy} + βE_{xy} -[v(v-1) (v-2) (y _{j'l'} - y _{i'l'}) - v(v-1) (y _{j'l} , + y _{j'} , + y _{j'} , + y _{j'}) -2r(v-2)y _{l,k'} + 4(v-1/y _{l,n}) ² /[2rv(v-1) v-2) [v ² -3v+2 (r-1) (v ² -v-1)]
Reciprocals	$v(v-1)/2$	r_{yy} + $\hat{\beta}E_{xy}$ + $[2y_{}-2ry_{k'} - v(v-1)(y_{1T} - y_{1T'})]^2/2rv(v-1)[(v(v-1) + 2(r-1)(v^2-v-1))]$
Error	$(r-1)(v^2-v-1)-1$	$E_{vv} - \beta E_{xv}$

Method II

$$
R_{yy} = 2\Sigma^{y^{2}} \cdot k \cdot k / v(v+1) - 2y^{2} \cdot \cdot \cdot / rv(v+1), \quad C_{yy} = \Sigma y^{2}{}_{ij} / r - 2y^{2} \cdot \cdot \cdot / rv(v+1)
$$

\n
$$
G_{yy} = [\Sigma (y_{i.} + y_{ii.})^{2} - 4y^{2} \cdot \cdot \cdot / v] / r (v+2),
$$

\n
$$
S_{yy} = \Sigma y^{2}{}_{ij} / r - \Sigma (y_{i.} + y_{ii.})^{2} / r (v+2) + 2y^{2} \cdot \cdot \cdot / rv(v+1) (v+2)
$$

\n
$$
Eyy = \Sigma y^{2}{}_{ijk} - \Sigma y^{2}{}_{ij} / r - 2 \Sigma y^{2}{}_{i.k} / v(v+1) + 2y^{2} \cdot \cdot \cdot / rv(v+1)
$$
\n(7)

Method III

$$
R_{yy} = \Sigma y^2_{\dots k} / v(v-1) - y^2_{\dots / rv}(v-1), \qquad C_{yy} = \Sigma y^2_{ij} / r - y^2_{\dots / rv}(v-1)
$$

\n
$$
G_{yy} = \Sigma (y_{i\dots} + y_{i\cdot})^2 / 2r(v-2) - 2y^2_{\dots / rv}(v-2), \qquad r_{yy} = \Sigma (y_{ij\dots} - y_{ji\cdot})^2 / 2r
$$

\n
$$
S_{yy} = \Sigma (y_{ij\cdot} + y_{ji\cdot})^2 / 2r - \Sigma (y_{i\dots} + y_{\cdot j\cdot})^2 / r(v-2) + y^2_{\dots / rv}(v-1) (v-2)
$$

\n
$$
E_{yy} = \Sigma y^2_{\cdot ijk} - \Sigma y^2_{\cdot ijk} / r - \Sigma y^2_{\dots k} / v(v-1) + y^2_{\dots / rv}(v-1)
$$
\n(8)

The terms like Ryy, *Cyy, G yy,* Syy' etc. are same as used for computing sums of squares in the analysis of variance model and the terms other than these are adjustment terms. The additional adjustment terms can easily be obtained by simply putting the totals from the diallel table.

 $\bar{1}$

February, 1997] Missing Plot Technique in Diallel Analysis

Table 5. ANOVA table when two observations are missing

In is v^2 for Method I, $v(v+1)/2$ for Method II, and (v^2-v) for Method III.

"Sca d.f. will be $v(v-3)/2$ for Method III.

"'Not applicable for Method 11.

The variances due to the estimates for various parameters are given below:

Method I

$$
var\left(\hat{g}_i\right) = \left[(v-1) + 2 \left(a_i \right)^{(g)} \right]^2 / (v^2 - 1)(r-1) \right] \sigma^2 / 2rv^2
$$
\n
$$
var\left(\hat{s}_{ij}\right) = \left[(v^2 - 2v + 2 + 2 \left(a_{ij} \right)^{(s)} \right]^2 / (v^2 - 1) \left(r - 1 \right) \right] \sigma^2 / 2rv^2
$$
\n
$$
(i \neq j)
$$

(9)

var (s_{ii}) =
$$
[(v-1)^2 + (a_{ii}^{(s)})^2/(v^2-1)(r-1)] \sigma^2/2rv^2
$$

var (r_{ij}) = $[v1 + (a_{ij}^{(r)})^2 / 2 (r-1)(v^2-1)] \sigma^2/2rv^2$

Method II

$$
\text{var}(\hat{S}_{ij}) = [(\mathbf{v} - 1) + (a_i^{(g)})^2 (\mathbf{v} + 1) / (\mathbf{v} + 2) (\mathbf{r} - 1) (\mathbf{v}^2 + \mathbf{v} - 2)] \sigma^2 / \text{rv}(\mathbf{v} + 2)
$$
\n
$$
\text{var}(\hat{S}_{ij}) = [(\mathbf{v}^2 + \mathbf{v} + 2) + \mathbf{v} (a_{ij}^{(g)})^2 / (\mathbf{v} + 2) (\mathbf{r} - 1) (\mathbf{v}^2 + \mathbf{v} - 2)] \sigma^2 / \text{r} (\mathbf{v} + 1) (\mathbf{v} + 2)
$$
\n
$$
\text{var}(\hat{S}_{ij}) = \mathbf{v} [(\mathbf{v} - 1) + (a_{ij}^{(g)})^2 / (\mathbf{v} + 2) (\mathbf{r} - 1) (\mathbf{v}^2 + \mathbf{v} - 2)] \sigma^2 / \text{r} (\mathbf{v} + 1) (\mathbf{v} + 2) \tag{10}
$$

Method **III**

var
$$
(\hat{g}_i)
$$
 = $[(v-1)/2rv (v-2) + (a_i (g))^2 (v-1) / 4r (r-1) v (v-2)^2 (v^2 - v-1)] \sigma^2$
\nvar (\hat{s}_{ij}) = $[v-3)/2r (v-1) + (a_{ij} (s))^2 v / 4r (r-1) (v-1) (v-2)^2 (v^2 - v-1)] \sigma^2$
\nvar (\hat{r}_{ij}) = $[1/2r + (a_{ij} (r))^2 (v^2 - v) / 4r (r-1) (v^2 - v-1)] \sigma^2$ (11)

The variance of differences between effects may also be computed in a similar manner. The values of a_i (g), a_{ij} (s), a_{ij} (s) and a_{ij} (r) are to be substituted from Table 1 depending on the applicability in a particular case.

MODEL AND ANALYSIS WHEN TWO OBSERVATIONS ARE MISSING

The covariance technique for handling of missing data can be easily extended to the case when two or more observations are missing by introducing one new concomitant variate for each missing observation and then using the multiple covariance technique for analysis. In case of two missing observations, assume additional covariate z apart from x in the model under consideration. The z takes value 0 or 1 similar to x. The observation y corresponding to both the missing values are taken to be zero. For example, the model for Method I can be written as:

$$
y_{ijk} = \mu + g_i + g_j + s_{ij} + r_{ij} + b_k + \beta_1 (x_{ijk} - \bar{x}) + \beta_2 (z_{ijk} - \bar{z}) + e_{ijk}
$$

(i, j = 1, 2, 3 ... v; k = 1, 2 ... r) (12)

where the parameters of the model are same as in equation (1) and β_1 , β_2 , are the regression coefficients of y on x and z, respectively. The model for other two methods can be written in the same way.

When two observations are missing, they may belong to two crosses, two parents, or one to a parent and the other to a cross in Methods I and II, while in Method III, they will belong only to crosses as there are no parents in this case. In general, let us assume yi'j'k' and yutm be the two missing observations on $(i' \times j')$ th and $(u \times t)$ th crosses in k'-th and m-th blocks, respectively.

Table 8. Values of $T_{xx}(T_{zz})$ and T_{xy} for Method III

Component		When observation on a cross is missing
	$T_{xx}(T_{zz})$	T_{xy}
Crosses	$(v^2-v-1)/(v^2-v)r$	$y_{i'i'}/r-y(rv(v-1))$
Parental effects (gca)	$1/\nu r$	$[v(y_{i}'+y_{i}'+y_{i}'+y_{i}')+4y_{i}]/2rv(v-2)$
Parental interactions (sca)	$(v-3)/2r(v-1)$	$(y_{i'i'} + y_{i'i'})/2r-(y_{i'}.+y_{i'}.+y_{i'}.+y_{i'})/2r(v-2)$ $+y/r(v-1)(v-2)$
Reciprocal effects	1/2r	$(y_{i'i'-y_{i'i'}})$ 2r
Error	$(r-1)(v^2-v-1)/rv(v-1)$	$(y_{}-ry_{k'}-v(v-1)^2y_{i'j'})/rv(v-1)$

The estimates of parameters are obtained by the leastsquares method. For example, the least square estimates in Method I are obtained as under:

 $\hat{\mu}$ = y.../rv² \hat{g}_i = $[v(y_{i..} + y_{i.}) - 2y \ldots - 2 (a_{i (g)} \hat{\beta}_1 + c_{i (g)} \hat{\beta}_2)]/2rv^2$ 78 L. S. Kaushik et al. [Vol. 57, No. 1]

$$
\hat{s}_{ij} = [v^2 (y_{ij.} + y_{ji.}) - v(y_{i..} + y_{.i.} + y_{j..} + y_{.j.}) + 2y ... - (a_{ij}^{(s)} \hat{\beta}_1 + c_{ij}^{(s)} \hat{\beta}_2)]/2rv^2
$$
\n
$$
\hat{s}_{ii} = [v^2 y_{ij.} - v(y_{i..} + y_{.i.}) + y ... - (a_{ii}^{(s)} \hat{\beta}_1 + c_{ii}^{(s)} \hat{\beta}_2)]/rv^2
$$
\n
$$
\hat{r}_{ij} = [(y_{ij.} - y_{ji.}) - (a_{ij}^{(r)} \hat{\beta}_1 + c_{ij}^{(r)} \hat{\beta}_2)]/2r
$$
\n(13)

where $c_i^{(g)}$, $c_{ij}^{(s)}$, $c_{ii}^{(s)}$ and $c_{ij}^{(r)}$ will have the same respective values as $a_i^{(g)}$, $a_{ij}^{(s)}$, a_{ii} (s) and a_{ij} (r) given in last section. The a's are concerned with $y_{i' j' k'}$ -th missing observation while c's are concerned with y_{utm} -th missing observation. The estimates of parameters in the remaining two methods are also obtained in a similar manner.

The values taken by the combination of a's and c's on substituting in the respective formula (13) will give required estimates of various parameters. For instance, when $y_{ij'k'}$ -th and y_{utm} -th observations on two different crosses in k'-th and m-th blocks are missing in Method I, each of $a_i^{(g)}$ and $c_i^{(g)}$ will take two values and we will have four possible estimates of g_i corresponding to the four possible combinations of $a_i^{(g)}$ and $c_i^{(g)}$. For example,

$$
\hat{g}_i = (y_{i..} + y_{i..})/2vr - y_{...}/rv^2 - (v-2)(\hat{\beta}_1 + \hat{\beta}_2)/2rv^2
$$
 if both missing crosses pertain to
\nthe i-th line (i=i'=u)
\n= (y_{i..}+y_{i..})/2vr - y.../rv² - (v-2) $\hat{\beta}_1/2v^2r + \hat{\beta}_2/rv^2$ if i-th line is involved in first
\nmissing observation (i=i'+ u)
\n= (y_{i..}+y_{i..})/2vr - y.../rv² + $\hat{\beta}_1/v^2r + (v-2)\hat{\beta}_2/2rv^2$ if i-th line is involved in second
\nmissing observation (i=u≠ i')
\n= (y_{i..} + y_{i.})/2vr + y.../rv² + ($\hat{\beta}_1 + \hat{\beta}_2$)/rv² otherwise

For estimating s_{ij} , a_{ij} ^(s) and c_{ij} ^(s) each will take all values except -(v-1) which will form 9 combinations and will thus give 9 possible estimates of s_{ij} . Similarly there will be four estimates of s_{ii} and 9 of r_{ii} .

When $y_{i'j'k'}$ -th and y_{utm} th observations are missing in Method II, each a_i (g) and c_i (g) will have two values, which will make four combinations, and on substituting in extended form of formula (3) they will have four estimates of g_i . Similarly, there will be nine estimates of s_{ii} and four of s_{ii} . The estimates for Method III can also be enumerated in a similar manner.

Now let us consider the case where two observations on parents, say yi'j'k' and yutm (i' \neq u) are missing. In Method I, each of a_j (g) and c_i (g) will have the values v-1 and -1; a_{jj} (s) and c_{ii} (s) will have the values -(v-1) and -1; a_{ii} (s) and c_{ii} (s) will have the values (v-1)² and 1 and a_{ij} ^(r) and c_{ii} ^(r) will have the values zero. The combinations of a's and c's will give four estimates each of g_i , s_{ij} , s_{ij} and one estimate of r_{ij} . In Method II, there will be four estimates each of g_i , s_{ii} and s_{ii} .

The estimates of β_1 and β_2 for observations missing on two different crosses i.e. (i' x j')th and $(u \times t)$ th in k'-th and m-th blocks are obtained, in general, as:

$$
\hat{\beta}_1 = (E_{zz}E_{xy} - E_{xz}E_{zy})/(E_{xx}E_{zz} - E_{xz}^2)
$$

and

$$
\hat{\beta}_2 = (E_{xx}E_{zy} - E_{xz}E_{xy})/(E_{xx}E_{zz} - E_{xz}^2)
$$
 (14)

Where E_{zz} and E_{xx} are error sums of squares for z and x; E_{zy} and E_{xz} as error sums of products of z with y and x, respectively; and E_{xy} is the error sum of the products of x with y.

Here $E_{xx} = E_{zz} = (r-1) (n-1)/rn$, $E_{xy} = (y... - ry_{x-k'} - ny_{i'j'})/rn$ and $E_{zy} = (y... - ry_{nm} - ny_{ny'})/rn$ $ny_{ut.}$)/ rn. The values of E_{zx} is to be obtained from bottom of Table (9).

The sums of squares obtained by the method of fitting constants are presented in Table (5). In this table C_{yy}, S_{yy,} G_{yy} and R_{yy} are computed by the formulae (6) or (7) or (8) depending on the method used and β_1 and β_2 are obtained by using the equation (14). $\hat{\beta}_1$ ^(C), $\hat{\beta}_1$ ^(P), $\hat{\beta}_1$ ^(PI), $\hat{\beta}_1$ ^(r) and $\hat{\beta}_2$ ^(C), $\hat{\beta}_2$ ^(PI), $\hat{\beta}_2$ ^(PI), $\hat{\beta}_2$ ^(r)) are also obtained using equation (14) by replacing the error sum of squares and sum of products term with T (treatment) + error, where T is C (cross effect), or P (parental effect), or PI (Parental interaction effect), or r (reciprocal effect). While computing these the values of β' s, T_{xx} , T_{zz} , T_{xy} are to be obtained from Tables 6, 7 or 8, depending on their applicability. The value of T_{zy} will also be same as of T_{xy} with i' replaced by u, j' by t, and k' by m. The value of T_{xz} is to be obtained from Tables 9, 10 or 11, depending on the applicability of the case.

The variance of the estimates obtained here can be seen to be a simple extension of the case of one missing observation, i.e. in the formulae $(9) - (11)$, a term due to the second missing observation will get introduced in a manner similar to that of the first.

For example in Method I, the variance of estimates are obtained as under:

 $var(\hat{\mu}) = \sigma^2 / rv^2$ var $({}^{0}_{8i}) = [(v-1) + 2 (a_i^{(g)})^2 + c_j^{(g)})^2 / (v^2-1) (r-1)]\sigma^2 / 2rv^2$

80 L. S. *Kaushik et al.* [Vol. 57, No.1

Table 9. Values of T_{xz} for Method I

***Two different parents or crosses or parent and a cross with no common parent.

Notes: $Exz = -(r-1)/m$ if two observations missing in the same replication.

 $Exz = -(n-1)/rn$ if two observations missing on the same cross or parent.

 $Exz = 1/m$ otherwise (i.e. none of above).

Table 10. Values of T_{xz} for Method II

Table 11. Values of T_{xz} for Method III

February, 1997] *Missing Plot Technique in Diallel Analysis* 81

$$
\text{var}(\hat{s}_{ij}) = \left[(v^2 + 2v + 2) + 2 ((a_{ij}^{\text{(s)}})^2 + (c_{ij}^{\text{(s)}})^2) / (v^2 - 1) (r - 1) \right] \sigma^2 / 2rv^2
$$
\n
$$
\text{var}(\hat{s}_{ij}) = \left[(v - 1)^2 + ((a_{ij}^{\text{(s)}})^2 + (c_{ij}^{\text{(s)}})^2) / (v^2 - 1) (r - 1) \right] \sigma^2 / rv^2
$$
\n
$$
\text{var}(\hat{r}_{ij}) = \left[1 + ((a_{ij}^{\text{(r)}})^2 + (c_{ij}^{\text{(r)}})^2) v^2 / 2 (r - 1) (v^2 - 1) \right] \sigma^2 / 2r
$$

The values of a's and C's are substituted depending on the applicability of the case. Variances of the differences between estimates can also be derived in a similar manner. The variances of the estimates for Method 2 and Method 3 can also be obtained in a similar manner. Moreover, they can be easily obtained by symmetry. Even for more than two missing observations, the extension of the technique is quite simple and straight forward by introducing one new concomitant variate for such missing observation and then using the multiple covariance technique for analysis.

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