AN INTERCROSS FAMILY WITH AN UNUSUAL PROPERTY

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Received 8.ii.79

SUMMARY

The detection and estimation of linkage is described for a completely classified intercross from which one array is missing. Provided that the missing array involves a gamete of non-recombinant type, this kind of family is capable of yielding more information about the linkage value in the affected parent than a completely represented family.

1. INTRODUCTION

LAWRENCE, CORNISH and HAYWARD (1979) have shown that it is possible to obtain independent estimates of male and female recombination frequency from a completely classified intercross family in which three or four alleles are segregating at each of a pair of linked loci. In the present note we consider a variant of this design with a rather unexpected property. It is convenient to refer to the first intercross as a type 1 family and to the one to be considered here as a type 1A family.

Consider a cross between an individual of genotype A_1B_1/A_2B_2 used as female and another of genotype A_1B_1/A_3B_3 used as male, where A_1 , A_2 and A_3 are alleles at the first and B_1 , B_2 and B_3 are those at the second locus. Suppose that male gametes of genotype A_1B_1 either do not occur, because of complementary gametic lethality, or fail to function, because

TABLE 1

The expected composition of progeny produced by crossing an A_1B_1/A_2B_2 individual used as female with an A_1B_1/A_3B_3 individual used as a male parent when male gametes of genotype A_1B_1 fail to survive or function. The n_{ij} 's are the number observed in each class and $t_m = (1 + p_m)$. Because of the missing male array, the row totals are partial sums. The grand total, $n_{..}$ is written as n in the text

	(A_1B_1) (0)	A_1B_3 p_m/t_m	A_3B_1 p_m/t_m	A_3B_3 q_m/t_m	Row totals
A_1B_1 $q_f/2$	—	A_1B_1/A_1B_3 $q_f p_m/2t_m$ n_{12}	A_1B_1/A_3B_1 $q_f p_m/2t_m$ n_{13}	A_1B_1/A_3B_3 $q_f q_m/2t_m$ n_{14}	$q_f/2$ n_1
A_1B_2 $p_f/2$	—	A_1B_2/A_1B_3 $p_f p_m/2t_m$ n_{22}	A_1B_2/A_3B_1 $p_f p_m/2t_m$ n_{23}	A_1B_2/A_3B_3 $p_f q_m/2t_m$ n_{24}	$p_f/2$ n_2
A_2B_1 $p_f/2$	—	A_2B_1/A_1B_3 $p_f p_m/2t_m$ n_{32}	A_2B_1/A_3B_1 $p_f p_m/2t_m$ n_{33}	A_2B_1/A_3B_3 $p_f q_m/2t_m$ n_{34}	$p_f/2$ n_3
A_2B_2 $q_f/2$	—	A_2B_2/A_1B_3 $q_f p_m/2t_m$ n_{42}	A_2B_2/A_3B_1 $q_f p_m/2t_m$ n_{43}	A_2B_2/A_3B_3 $q_f q_m/2t_m$ n_{44}	$q_f/2$ n_4
Column totals	—	p_m/t_m n_2	p_m/t_m n_3	q_m/t_m n_4	1 $n_{..}$

of their incompatibility on stigmata which carry the same alleles. Then if p_f is the frequency of recombination on the female and p_m is the corresponding frequency on the male side of the cross and there are no further viability disturbances, the frequencies of the genotypes expected in the progeny are as shown in table 1 ($q_f = 1 - p_f$ and $q_m = 1 - p_m$). The actual or effective absence on the male side of one of the four gametic genotypes means, of course, that the frequencies of the surviving three have to be divided by $t_m = (1 + p_m)$ in order that they shall sum to unity; and that we now have 12 zygotic genotypes, rather than the 16 of the type 1 intercross in which all four kinds of male gamete participate in fertilisation. However, as in the latter, if there is no dominance at either locus, each genotype will produce a unique phenotype. The present family is thus an incompletely represented but completely classified intercross.

2. ESTIMATION OF MALE AND FEMALE LINKAGE

The test for linkage on the female side is the same as that for the type 1 family, namely:

$$\chi^2_{(1)} = (n_{1.} - n_{2.} - n_{3.} + n_{4.})^2 / n \\ = F^2 / n$$

where F is written for $(n_{1.} - n_{2.} - n_{3.} + n_{4.})$ and n is written for the $n_{..}$ of table 1. On the male side, however, the test for linkage now becomes:

$$\chi^2_{(1)} = (2n_{.4} - n_{.2} - n_{.3})^2 / 2n = M^2 / 2n$$

where M is written for $(2n_{.4} - n_{.2} - n_{.3})$.

The joint test for linkage and the test of heterogeneity between the male and female sides will be discussed after the estimation of the linkage values has been considered.

Turning to the estimation of the linkage values, p_f and p_m , for the female and male recombination fractions respectively, the logarithm of the likelihood is

$$L = [(n_{22} + n_{23} + n_{24}) + (n_{32} + n_{33} + n_{34})] \log p_f \\ + [(n_{12} + n_{13} + n_{14}) + (n_{42} + n_{43} + n_{44})] \log q_f \\ + [(n_{12} + n_{22} + n_{32} + n_{42}) + (n_{13} + n_{23} + n_{33} + n_{43})] \log p_m \\ + [(n_{14} + n_{24} + n_{34} + n_{44})] \log q_m \\ - n \log (1 + p_m) + \text{constants}$$

which on partial differentiation in respect of p_f and p_m gives as the two equations of estimation:

$$\frac{\partial L}{\partial p_f} = \frac{n_{2.} + n_{3.}}{p_f} - \frac{n_{1.} + n_{4.}}{q_f} = 0 \\ \frac{\partial L}{\partial p_m} = \frac{n_{.2} + n_{.3}}{p_m} - \frac{n_{.4}}{q_m} - \frac{n}{1 + p_m} = 0$$

The two estimates are thus

$$p_f = \frac{n_{2.} + n_{3.}}{n} \quad \text{and} \quad p_m = \frac{n_{.2} + n_{.3}}{n_{.4} + n}$$

We note that on the female side both the equation of estimation and the estimate are the same as those from the type 1 intercross, except that the row sums involve three rather than four cells. Both the equation of estimation and the estimate of recombination frequency on the male side, on the other hand, are different from the previous case. Despite this, however, the estimates of p_f and p_m are still independent: indeed the log likelihood is composed of two completely separable parts, one dependent solely on p_f and yielding the estimate of this parameter and the other similarly dependent solely on p_m .

The amounts of information about the estimates yielded by a family of the present type are:

$$I_{p_f} = \frac{n}{p_f q_f} \quad \text{and} \quad I_{p_m} = \frac{2n}{(1+p_m)^2 p_m q_m}$$

Again, as with the estimate itself, the amount of information about recombination on the female side of the cross is the same as that for the previous design. The corresponding statistics on the male side, however, take a novel form.

3. THE EFFICIENCY OF THE MALE ESTIMATE

Now in the completely classified cross with 16 classes, the amount of information yielded by an individual in respect of p_m is

$$i_{p_m} = \frac{I_{p_m}}{n} = \frac{1}{p_m q_m}$$

In the present case this quantity is

$$i'_{p_m} = \frac{I'_{p_m}}{n} = \frac{2}{(1+p_m)^2 p_m q_m}$$

The relative efficiency of the type 1A compared with the type 1 family in respect of the estimation of recombination on the male side is thus:

$$\text{Efficiency \%} = i'_{p_m}/i_{p_m} = \frac{2}{(1+p_m)^2} \times 100$$

Since this efficiency depends on p_m , it is convenient to plot values of the former against the latter in the form of a graph (figure 1).

It is quite clear that for values of p_m less than $\sqrt{2}-1 = 0.41$, the present design is more efficient than the previous one. Indeed with very tight linkage, this incomplete cross is nearly twice as efficient as the type 1 cross. This is, of course, a rather unexpected outcome since in general incomplete families are less informative than complete ones. However, it is worth emphasising that this gain in information is obtained only if the appropriate male array is missing. Thus in the present case we have assumed for the purpose of illustration that the linkage between A_1 and B_1 is in the coupling phase and that male gametes of genotype $A_1 B_1$ fail to function. Gametes of this genotype are, of course, non-recombinant with respect to the alleles carried by the male parent. If this assumption about the phase of linkage is

invalid, the present design is less efficient than the previous one. Thus if linkage is in the repulsion phase ($p > 0.5$ in the graph), the efficiency of the type 1A design can fall to no more than 50 per cent of that of the type 1 design. In practice, it may be possible to circumvent this difficulty by rearranging the cross so as to ensure that the missing array concerns a male

FIG. 1.—The efficiency (%) of a type 1A family relative to that of a completely classified intercross (type 1) in respect of the estimation of the male linkage parameter p_m .

gamete of non-recombinant genotype; for example, the cross $A_1B_3/A_2B_2 \times A_1B_3/A_3B_1$ would fulfill this objective. If, however, this cannot be done or the phase of linkage is not known in the parents it is clearly safer to use the type 1 design.

The second point worth making about this design is that it should not be supposed that it is possible to obtain a gain in efficiency by discarding part of the data from a complete family. Thus, suppose that we score n individuals in a type 1 and n' individuals in a type 1A intercross.

Then

$$I_{p_m} = \frac{n}{p_m q_m} \quad \text{and} \quad I'_{p_m} = \frac{2n'}{(1+p_m)^2 p_m q_m}$$

so that

$$I'_{p_m}/I_p = \frac{2}{(1+p_m)^2} \frac{n'}{n}$$

Supposed now that we discard the A_1B_1 male array from a type 1 family in which it occurs with a frequency of $q_m/2$. Then the total number of individuals in this simulated type 1A family is

$$n' = n(1 - q_m/2) = n(1 + p_m)/2$$

so that

$$I'_{p_m}/I_{p_m} = \frac{1}{1+p_m}$$

Since $1/(1+p_m)$ is less than unity for all values of p_m greater than zero, it is clearly impossible to obtain a gain in information about p_m by discarding the A_1B_1 male array from a complete intercross family.

It should be noted that in theory a similar gain in efficiency could be obtained in a backcross family, though it is difficult to imagine the circumstances in which it would be possible in practice to construct the cross.

4. THE JOINT ESTIMATE AND DETECTION OF LINKAGE

When there is no reason to suppose that p_f and p_m differ in value a joint estimate of p may be obtained. Substituting p for both p_f and p_m in the log likelihood expression, and differentiating in respect of p gives as the equation of estimation

$$\frac{dL}{dp} = \frac{n_2 + n_3}{p} - \frac{n_1 + n_4}{q} + \frac{n_2 + n_3}{p} - \frac{n_4}{q} - \frac{n}{1+p} = 0$$

in which the first two terms can be recognised as stemming from the female side and the last three from the male. This equation can be reduced to

$$(\alpha + \beta)p^2 + (\alpha + 3\beta) - 2\alpha = 0$$

where $\alpha = (n_2 + n_3 + n_{2.2} + n_{3.3})$ and $\beta = (n_1 + n_4 + n_{1.4})$ with $\alpha + \beta = 2n$. The appropriate root of this quadratic equation provides the joint estimate, p .

The amount of information about p is given by the sum of the amounts for p_f and p_m , where they are estimated separately, with, of course, p substituted for p_f and p_m .

Thus

$$I_{p_f} + I_{p_m} = n/p_f q_f + 2n/p_m q_m (1 + p_m)^2$$

giving

$$I_p = \frac{n}{pq} \left[1 + \frac{2}{(1+p)^2} \right]$$

The joint expression, dL/dp , can also be used to derive the two χ^2 's which provide respectively the joint test of significance for linkage (the

deviation $\chi^2_{(1)}$) and the test of agreement between the male and female sides (the heterogeneity $\chi^2_{(1)}$). The null hypothesis of no linkage requires that $p = q = \frac{1}{2}$. Substituting these values in dL/dp gives

$$D = \frac{2}{3}[3(n_{2.} + n_{3.} - n_{1.} - n_{4.}) + 2(n_{.2} + n_{.3} - 2n_{.4})] = \frac{2}{3}(3F + 2M)$$

where D is the departure from the value 0 expected on the null hypothesis—a departure which must arise by sampling variation alone if the null hypothesis is true. The sampling variance of D is I_p which when $p = \frac{1}{2}$ is $17 \times 4n/9$. D^2/I is distributed as a $\chi^2_{(1)}$ giving us the joint test of significance for linkage

$$\chi^2_{(1)} = \frac{4}{9}(3F + 2M)^2 \bigg/ \frac{17 \times 4n}{9} = (3F + 2M)^2/17n$$

The test of significance for the heterogeneity between the male and female sides in respect of the evidence they provide for linkage is similarly given by

$$\chi^2_{(1)} = (3F - 2M)^2/17n$$

Adding these two $\chi^2_{(1)}$ yields $\chi^2_{(2)} = 2[(3F)^2 + (2M)^2]/17n$, which is not identical with the $\chi^2_{(2)} = \frac{F^2}{n} + \frac{M^2}{2n} = \frac{2F^2 + M^2}{2n}$ obtained by summing the two χ^2 's found on page 124 for the two sides taken separately. This sum would, in fact, correspond to a deviation $\chi^2_{(1)} = (\sqrt{2F + M})^2/4n$ and a heterogeneity $\chi^2_{(1)} = (\sqrt{2F - M})^2/4n$.

The difference between this $\chi^2_{(2)}$ and that obtained by the maximum likelihood approach is, however,

$$\begin{aligned} \frac{1}{2n}(2F^2 + M^2) - \frac{2}{17n}(9F^2 + 4M^2) &= \frac{1}{34n}(M^2 - 2F^2) \\ &= \frac{1}{34n}(M - \sqrt{2F})(M + \sqrt{2F}) \end{aligned}$$

which will vanish when $F = M = 0$ (i.e. when the expectations from the null hypothesis are exactly realised) or when $M = \pm \sqrt{2F}$ (i.e. when the weighted deviations of the two sides exactly balance). Thus when the hypothesis of no linkage is acceptable the discrepancy between the χ^2 's obtained from the two approaches springs from sampling variation. The maximum likelihood approach is obviously to be preferred but the discrepancy will in any case be small and the alternative χ^2 's can be used without fear of serious misjudgment. If, however, the deviation χ^2 is significant, thus rejecting the hypothesis of no linkage, the heterogeneity χ^2 found using this hypothesis will inevitably be untrustworthy and could be misleading. In such circumstances heterogeneity should be tested using the value of p estimated from the data, using the method described by Mather (1938, 1951, section 18).

5. REFERENCES

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