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Location of X-linked polygenic effects causing sterility in male hybrids of *Drosophila simulans* and *D. mauritiana*

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There is general agreement that hybrid male sterility in *Drosophila* is caused by changes at several (perhaps many) factors, most of them located on the X chromosome. These factors have been generally considered as major genes, each one of them able to bring about sterility by itself. However, the evidence on this last point is not conclusive. In principle, the possibility that they correspond to located polygenic effects instead of genes with a large effect cannot be excluded. This paper shows that some of the factors that cause male sterility in *D. simulans*/*D. mauritiana* hybrids, located by recombination on the X chromosome, are indeed 'effective factors', or located polygenic effects. Some of the consequences of this finding are explored.

Keywords: *Drosophila*, male hybrids, polygenes, recombination, sterility.

Introduction

Hybrid males of *Drosophila simulans* and *D. mauritiana* are always sterile, in spite of some recent claims to the contrary (Goulielmos & Alahiotis, 1989). Genetic analysis of this hybrid sterility has traced the genes with the greatest effect to the X chromosome (Coyne & Charlesworth, 1989), in agreement with most genetic studies of postzygotic reproductive isolation in other species (reviewed in Charlesworth *et al.*, 1987; Coyne & Orr, 1988). In the case of *D. simulans* and *D. mauritiana*, three morphological markers (*yellow–white, miniature*, and *forked*) were used to map, by measuring recombination frequencies, the location of X-chromosome genes causing the sterility of their male hybrids. It was concluded (Coyne & Charlesworth, 1989) that one locus with large effects on sterility was tightly linked to each one of the morphological markers, and, consequently, that at least three X-linked major genes had diverged between the two species. This represented the maximum genetic divergence detectable with the three, randomly chosen, markers, and suggested that many other 'sterility' loci were present on the X chromosome. This last point is quite reasonable, and it may be agreed that hybrid male sterility is caused by changes at several (and perhaps many) genes. On the contrary, the question of the effect of each of these genes is far from being a settled matter. In so far as the effect on the phenotype is concerned, these genes might, in principle, be of two kinds: either major genes, each one able to bring about sterility of the hybrids by itself, or minor genes, whose individual effects would be relatively small but cumulative, and would bring about sterility only when combined in sufficient number (polygenic combination). By definition, then, it would not be possible to study polygenes individually, because their segregation would be unrecognized phenotypically. According to this last view, hybrid sterility should be considered to be a quasi-continuous or threshold-character because males would fit in either of two categories, fertiles or steriles, as a result of the combined effects of multiple non-allelic minor genes. In contrast with this view, most contemporary authors favour the former possibility, and consider that the interspecific sterility factors they have found in *Drosophila* correspond to genes with large effect, easily mappable by the conventional method of recombination with chromosome markers (Charlesworth *et al.*, 1987; Coyne & Charlesworth, 1989; Orr, 1989a, b; Zouros *et al.*, 1988). Using a different method, however, Naveira & Fontdevila (1986, 1991a, b) claim to have found what appears to be mainly a polygenic basis for sterility in the hybrids between two other *Drosophila* species, *D. buzzatii* and *D. koepferae*, at least as far as the autosomes are concerned. It is possible that these apparently conflicting observations

are the reflection of true differences in the genetic architecture of the reproductive isolating barriers among the species concerned but it seems worth while investigating whether it could be simply a question of different interpretations of essentially similar datasets. Is it possible that the same data could simply be viewed differently?

Shrimpton & Robertson (1988a, b) recently discussed the difficulties that must be faced by any attempt to map the genetic factors that determine any phenotype, when several factors are involved. According to them, citing Mather & Jinks (1971), a factor may correspond to a polygenic effect, genetically located by recombination, granted that subsequent recombination may further divide such a factor, now called an 'effective factor', into still smaller but linked effects. This offers the key to solve the problem. An experiment needs to be designed to test whether it is possible to subdivide by recombination a factor of hybrid sterility into smaller, linked effects. If such a subdivision is feasible, that factor should no longer be regarded as a gene with large effects on sterility but rather as a located polygenic effect, produced by the combined action of a polygenic set. In the present paper the results are reported of such an experiment, performed with the hybrids of *D. simulans* and *D. mauritiana*, and some of the consequences that these results may have are discussed.

Materials and methods

D. simulans and *D. mauritiana* can be crossed to yield fertile F_1 females and sterile F_1 males. Female hybrids can then be backcrossed to males of either parental species, yielding backcross males that are either fertile or sterile.

Males of *D. mauritiana* were crossed to female *D. simulans* homozygous for three recessive X-linked marker alleles (Fig. 1). The heteroygous hybrid females were then backcrossed to males from the *D. simulans* multiple marker stock, yielding backcross males segregating for the marker and the wild-type allele at each locus. The fertility of these males was tested by crossing those of the same phenotype with females of the *D. simulans* multiple-marker stock. Backcross males with a mutant marker carry an X-linked segment from *D. simulans* and can be fertile, while those with a wild-type allele carry a segment from *D. mauritiana* and are generally sterile (Coyne, 1984; Coyne & Kreitman, 1986; Coyne & Charlesworth, 1986, 1989). Heterozygous, wild-type females for any of the marked loci were selected among the backcross progeny, and females of the same phenotype were backcrossed again to males from the *D. simulans* marker stock. These backcrosses were repeated for a number of generations, with wild-type males for any of the marked loci being tested for fertility each generation. This crossing scheme allows a gradual reduction in linkage between each of the *D. mauritiana*-derived wild-type alleles and any sterility factors. In this way, several fertile wild-type backcross males for some of the marked loci were obtained. Upon crossing each one with females from the *D. simulans* marker stock, heterozygous, wild-type females carrying exactly the same segment from *D. mauritiana* could be obtained, which were crossed with their mutant brothers. In the next generation, wild-type females were selected and crossed with their sibling wild-type males, and in two successive generations, fixed hybrid stocks could be established (Fig. 1).

The three *D. simulans* mutations used in this study were *yellow* (*y*, 1–0.0); *white* (*w*, 1–4.1); and *forked* (*f*, 1–56.0). Mutant stocks were provided by the Mid-American *Drosophila* Stock Center, Bowling Green, Ohio.

D. mauritiana were provided by Dr J. F. McDonald (UGA) from a collection by Dr J. David (CNRS, France).

This crossing scheme finally led to the production of four hybrid stocks carrying alleles $y^{\pm} w^{+}$, three hybrid stocks carrying allele f^{+}, and two stocks carrying allele y^{+}. The other marker loci in these stocks were fixed for the mutant, *D. simulans* alleles. Each one of these strains was originally associated with one or several independent crossover events in the meiosis of a heterozygous, hybrid female backcrossed to *D. simulans*. Therefore, the chromosome segment from *D. mauritiana* actually linked to a given wild-type allele, or alleles, is not the same in the different replicates. In addition, two other stocks carrying allele w^{+} were derived from two of the former strains carrying alleles $y^{+} w^{+}$, by subsequent recombination with the *D. simulans* marker stock, and therefore the segments from *D. mauritiana* carried in them must be smaller, crossover products of those carried in the $y^{+} w^{+}$ strains.

Once all the fixed hybrid stocks were established, crosses were performed among them (Table 1). In the F_2, recombinant males that carried the wild-type alleles from both grandparental hybrid strains could be obtained. These hybrid males, produced by recombination in trans-heterozygous F_1 females, should carry a new combination of the segments from *D. mauritiana* carried in the grandparental hybrid stocks. The fertility of these males was scored by observing sperm motility (Coyne, 1984; Coyne & Charlesworth, 1986, 1989). Testes were removed from virgin males held for 3 days at 24°C, squashed, and inspected under a phase-contrast microscope. Males lacking sperm or possess-

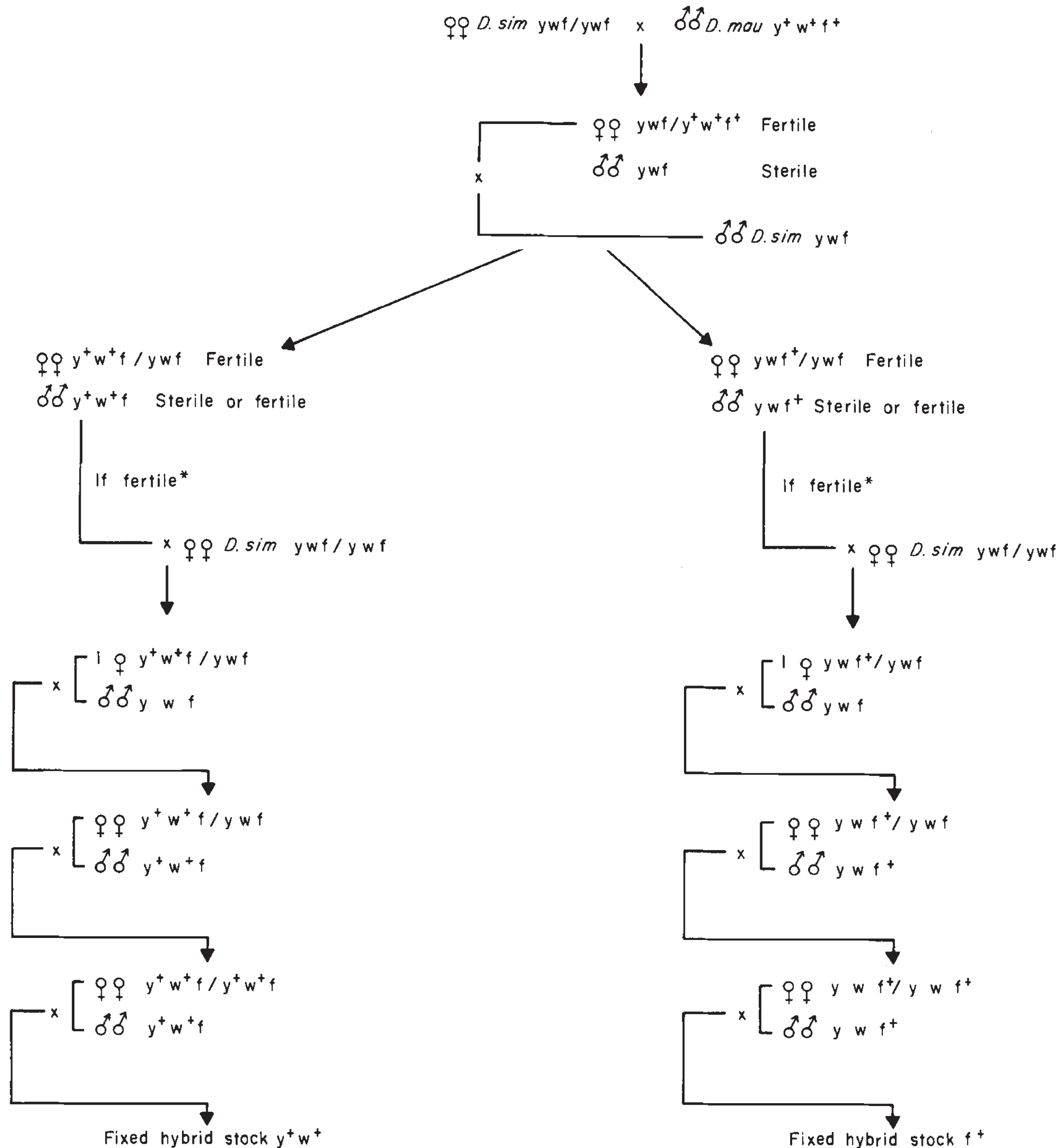

Fig. 1 Crossing scheme for the production of fixed hybrid stocks ($y^+ w^+$ and f^+, in this case). If fertile males are not found in *, backcrossing of hybrid females is repeated as many times as necessary.

ing only immotile sperm were scored as 'sterile', and those with at least one motile sperm as 'fertile'. The fertility of the grandparental-type males produced in these crosses (carrying the wild-type alleles from either of the two grandparental hybrid strains) was also scored.

All flies were kept in 2 × 8 cm glass vials at 24°C. The fly medium used was the standard mixture (cornmeal, yeast, agar) of this laboratory.

Results

Male sterility frequencies were first determined in the recipient *D. simulans* marker stock (100 males), and in the different fixed hybrid strains produced by introgression of *D. mauritiana* alleles in it (40 males in each strain). As expected, sterility frequencies in these hybrid strains were very low, and not significantly different from the frequency observed in *D. simulans* (homogeneity $\chi^2 = 6.94$, 11 d.f., $P \approx 0.80$). All these frequencies were therefore pooled to give a combined estimate of the frequency of background sterility in the recipient *D. simulans* stock, namely 5/540 = 0.9 per cent. Apparently, sterility loci, once linked to *D. mauritiana* alleles, have been lost in all the fixed hybrid stocks. Nevertheless, this does not mean that hybrid males in these stocks are as fertile as pure *D. simulans* or *D. mauritiana* males. As described in the Materials

Table 1 Sterility frequencies in different types of F_2 male offspring from crosses between fixed hybrid stocks. (a) Wild-type markers of the grandmother hybrid stock. (b) Wild-type markers of the grandfather hybrid stock. Sterility frequencies in each type of F_2 male offspring were tested for deviations from the background sterility of the recipient *D. simulans* strain (5/540 = 0.9%) by χ^2 tests. A sequential Bonferroni test was used to judge the significance at the 1 per cent level of the different tests corresponding to either (a + b), (a), or (b) F_2 males

F_2 males											
						Grandparental types					
Grandparental hybrid stocks			Recombinant wild-type (a + b)			(a)			(b)		
(a)	cross	(b)	Sterile/Total	Percentage	Significance	Sterile/Total	Percentage	Significance	Sterile/Total	Percentage	Significance
y^+w^+-1	×	f^+-1	27/38	71	s	8/36	22	s	4/22	18	s
y^+w^+-1	×	f^+-2	16/21	76	s	4/20	20	s	0/20	0	ns
y^+w^+-1	×	f^+-3	7/20	35	s	2/19	11	ns	3/14	21	s
y^+w^+-2	×	f^+-1	6/25	24	s	0/24	0	ns	1/20	5	ns
y^+w^+-2	×	f^+-2	0/23	0	ns	1/24	4	ns	0/22	0	ns
y^+w^+-2	×	f^+-3	2/20	10	ns	0/20	0	ns	0/20	0	ns
y^+w^+-3	×	f^+-1	13/36	36	s	5/35	14	s	0/16	0	ns
y^+w^+-3	×	f^+-2	11/27	41	s	0/27	0	ns	0/25	0	ns
y^+w^+-3	×	f^+-3	4/41	10	s	0/27	0	ns	0/20	0	ns
y^+w^+-4	×	f^+-1	10/20	50	s	3/20	15	s	1/20	5	ns
y^+w^+-4	×	f^+-2	11/20	55	s	2/20	10	ns	0/20	0	ns
y^+w^+-4	×	f^+-3	12/20	60	s	2/20	10	ns	0/20	0	ns
w^+-1	×	f^+-1	20/33	61	s	0/25	0	ns	0/18	0	ns
w^+-1	×	f^+-2	7/21	33	s	0/20	0	ns	0/18	0	ns
w^+-1	×	f^+-3	0/21	0	ns	0/23	0	ns	0/11	0	ns
w^+-2	×	f^+-1	3/22	14	s	1/19	5	ns	0/20	0	ns
w^+-2	×	f^+-2	0/20	0	ns	0/20	0	ns	0/20	0	ns
w^+-2	×	f^+-3	2/39	5	ns	0/25	0	ns	0/23	0	ns
y^+-5	×	w^+-1	0/2	0	–	0/20	0	ns	1/22	5	ns
y^+-5	×	w^+-2	0/1	0	–	0/6	0	–	0/3	0	–
y^+-6	×	w^+-1	0/2	0	–	0/20	0	ns	0/20	0	ns
y^+-6	×	w^+-2	0/1	0	–	0/20	0	ns	0/20	0	ns
y^+-6	×	f^+-1	1/20	5	ns	0/20	0	ns	0/20	0	ns
y^+-6	×	f^+-2	1/18	6	ns	0/20	0	ns	0/20	0	ns
y^+-6	×	f^+-3	0/20	0	ns	0/20	0	ns	0/20	0	ns

and Methods section, a male is scored as 'fertile' when at least one sperm is motile. Actually, two males may have been equally scored as 'fertile' and still differ significantly in their amounts of motile sperm (i.e. potential fertility or fecundity). No detailed analysis of this was performed but it is something that should be borne in mind.

Fixed hybrid strains carrying different wild-type alleles from *D. mauritiana* were crossed in pairs, as indicated in Table 1. F_2 males corresponding to either wild-type recombinanat or parental- (grandparental) type gametes were identified and scored for fertility. Sterility frequencies in these different types of male were tested for deviations from background sterility of the recipient *D. simulans* strain by $2 \times 2\chi^2$-tests. To reduce the probability of incorrectly rejecting one or more true null hypotheses (type I errors), a sequential Bonferroni test (Rice, 1989) was used to judge the significance at the 1 per cent level of the different component tests, corresponding to wild-type recombinant (21 simultaneous tests), and either parental type F_2 males (24 simultaneous tests each). The results are shown in Table 1. It can be clearly seen that many of the crosses gave rise to significant frequencies of sterility in F_2 males but most particularly among the wild-type recombinants, which combine the wild-type alleles from both the hybrid stocks involved in the cross. The only exceptions to this pattern are the crosses that involve the *yellow* marker, which produced mostly F_2 fertile males, no matter which strain they were crossed with. On the other hand, percentages of sterile males differed substantially from one cross to another, even when keeping one stock constant. For example, when sterility frequencies are compared among crosses involving the same grandmother hybrid strain by a χ^2 homogeneity test, significant differences are found among recombinant wild-type F_2 males from the crosses of the different f^+ strains with $y^{\pm} w^+ - 1$ ($P<0.01$), $y^+ w^+ - 2$ ($P<0.05$), $y^+ w^+ - 3$ ($P<0.01$), and $w^+ - 1$ ($P<0.01$); and among the males of one of the grandparental-types from the crosses with $y^+ w^+ - 3$ ($P<0.05$). All these results demonstrate that the fixed hybrid strains obtained in the first part of this experiment had not entirely lost the factors which can cause hybrid male sterility. Some sterility factors must still be linked to the wild-type alleles from *D. mauritiana* introgressed into *D. simulans* in the different hybrid strains, factors that are apparently able to interact and bring about sterility again when joined on the same X chromosome by recombination.

It is also useful to compare the results of strains $y^+ w^+ - 1$ and $y^+ w^+ - 2$, with those from their derived strains, $w^+ - 1$ and $w^+ - 2$, respectively. The only genetic difference between these two sets of strains is a *D. mauritiana* segment marked by the y^+ allele, which must always be very small (the distance between y and w loci is 4.1 map units). Two other segments of this kind ($y^+ - 5$ and $y^+ - 6$) were unable to bring about sterility when combined with segments marked by w^+ or f^+ *D. mauritiana* alleles (Table 1). This does not mean however that y^+ segments are entirely devoid of sterility factors. Table 1 shows that sterility frequencies in F_2 males from the crosses of f^+ strains with $w^+ - 1$ or $w^+ - 2$ are generally smaller than the corresponding sterility frequencies observed in the crosses with $y^+ w^+ - 1$ or $y^+ w^+ - 2$, the differences proving to be significant in the case of $y^+ w^+ - 1$ versus w^+ ($P<0.01$, according to a simple sign test). Therefore, y^+ segments may indeed contain hybrid male sterility factors, although probably in a very small number, in correspondence with their small size.

Discussion

In a recent paper, Naveira & Fontdevila (1991a) have shown that two kinds of sterility, either genic or chromosomal, may be found in hybrid males between two species of the *Drosophila buzzatii* cluster. Genic sterility is brought about by genes of large effect (major genes), whereas chromosomal sterility has a polygenic basis, being produced by the cumulative action of multiple factors of minor effect (Naveira & Fontdevila, 1991b). In the autosomes of these species, most sterility factors corresponded to located polygenic effects, whereas the evidence concerning the X chromosome was inconclusive because none of the investigated X chromosome segments, which represented at least 1 per cent of the haploid polytene karyotype, allowed hybrid male fertility. This result indicates that there are several factors of hybrid sterility on the X, but nothing about their nature because the length of introgressed segments could not be further reduced by subsequent recombination. There is, apparently, a lower limit to this length, due to the inhibition of crossing-over by the characteristic asynapsis of homologous chromosomes in interspecific hybrids (Evgenev, 1971). It seemed reasonable, therefore, to investigate two other, more closely related species of *Drosophila*, where the role of the X chromosome could be assessed. *D. simulans* and *D. mauritiana* offered such an opportunity. Their F_1 hybrid males are always sterile but fertile backcross males with X-linked markers have been recovered by other authors, who investigated in great detail the genetic basis of this isolating barrier (see the introduction for references).

It is advisable, first, to emphasize that recent claims on the exceptional fertility of F_1 *D. simulans*/*D. mauritiana* hybrid males (Goulielmos & Alahiotis,

1989) are open to question. On the one hand, the putative hybrid stocks are no longer available to other workers, and on the other, the interpretation of electrophoretic patterns by the authors, which constitutes the basis for their claim, is highly questionable because the homogenates analysed were of groups of flies, rather than single flies. Therefore, it is enough to postulate the presence of one or several non-virgin females in the crosses to explain the observed results (mixed stocks of hybrid and pure flies). In conclusion, and in agreement with the rest of the literature, we must still consider that F_1 hybrid males between *D. simulans* and *D. mauritiana* are always sterile, no matter what strains are used for hybridizaition.

Coyne & Charlesworth (1986, 1989) located, by recombination, three sections of the X chromosome of *D. mauritiana* that, in their interpretation, contained genes with substantial effects on hybrid male sterility (major genes). If this is the case, by combining in the same male two sections from *D. mauritiana* that separately do not produce sterility, on would expect to obtain always fertile males, because under this hypothesis a male would be fertile whenever it had lost the sterility factors initially linked to the *D. mauritiana* marker alleles. The results presented in this paper constitute a clear rebuttal of this hypothesis. It is shown that at least two of the located sterility effects may be further subdivided by recombination into still smaller, linked effects, which constitute a standard test for the presence of polygenes. Sterility, a threshold character, would then be brought about whenever the introgressed chromosome section from *D. mauritiana* was large enough to contain a critical number of interacting polygenes. This hypothesis provides a satisfactory explanation for all the findings of the experiments described in this paper.

1 The reconstitution of hybrid sterility after recombining in the same male two marked chromosome sections from *D. mauritiana* that, separately, allow fertiliy. The combination of the two sections brings together a number of polygenes sufficient to cause sterility but each separate section harbours a lower-than-critical number.

2 The occasional appearance of sterile males among grandparental-type hybrid males. Undetected recombination may increase the number of polygenic sterility factors, thus linking to the grandparental-type X chromosome a higher-than-critical number of factors, which result in hybrid male sterility.

3 The contribution to hybrid male sterility of the small section marked by the y^+ allele. Polygenic sterility factors must be very abundant and distributed all over the X chromosome. Thus, even a relatively small chromosome section, such as that linked to y^+, may make a significant contribution when combined with a section containing a near-critical number of polygenes (the number that corresponds to the threshold between fertility and sterility).

4 The high heterogeneity in sterility frequencies among crosses involving one given strain. The extent and localization of *D. mauritiana* segments, and, consequently, of polygenic sterility sets on the X chromosome of the hybrid, is largely a matter of chance. Each hybrid strain is effectively unique in its combination of *D. mauritiana* and *D. simulans* chromosome sections, and thus gives rise to unique interactions with the other strains. However, some general characteristics of the strains can be deduced from the results. For example, strain $y^+ w^+ - 1$ should carry the largest introgressed segment because it gives rise to the highest sterility frequencies in Table 1. Strain $y^+ w^+ - 2$ should carry a relatively large introgressed segment between the f locus and the centromere, because it shows very low sterility frequencies, or no sterility at all, after recombination with f^+ chromosomes. Among f^+ strains, $f^+ - 3$ should carry the smallest introgressed segment but most of this segment should correspond to the proximal (centromeric) region.

In conclusion, the results reported in this paper, which point to the presence of several X-linked 'effective factors' of sterility in hybrids between *D. simulans* and *D. mauritiana*, are entirely consistent with the results obtained for autosomes in the *buzzatii* cluster (Naveira & Fontdevila, 1986, 1991a, 1991b). Furthermore, it is quite probable that many of the hybrid sterility factors, so far reported in the literature (Coyne, 1984; Coyne & Kreitman, 1986; Coyne & Charlesworth, 1989; Dobzhansky, 1936; Orr, 1987, 1989a, b; Orr & Coyne, 1989; Pontecorvo, 1943; Vigneault & Zouros, 1986; Zouros *et al.*, 1988), do not correspond to genes with large effects but to 'effective factors' instead. One first problem now is to design methods to estimate the mean and variance of the critical size for hybrid sterility (the size, in map units, that an introgressed chromosome segment should have to bring about sterility), which should correspond to a minimum number of introgressed polygenes from *D. mauritiana*. A second problem is the estimation of the relative importance of these minor sterility factors in the X chromosome, as compared with major genes (are there any major genes of hybrid sterility at all?). A third, and final problem, is the determination of the nature of these polygenes, and of how their interactions with autosomal and/or Y-linked alleles from *D. simulans* bring about sterility.

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References

CHARLESWORTH, B., COYNE, J. A. AND BARTON, N. H. 1987. The relative rates of evolution of sex chromosomes and autosomes. *Am. Nat.*, **130**, 113–146.

COYNE, J. A. 1984. Genetic basis of male sterility in hybrids between two closely related species of *Drosophila. Proc. Natl. Acad. Sci., U.S.A.* **81**, 4444–4447.

COYNE, J. A. AND CHARLESWORTH, B. 1986. Location of and X-linked factor causing sterility in male hybrids of *Drosophila simulans* and *D. mauritiana. Heredity*, **57**, 243–246.

COYNE, J. A. AND CHARLESWORTH, B. 1989. Genetic analysis of X-linked sterility in hybrids between three sibling species of *Drosophila. Heredity*, **62**, 97–106.

COYNE, J. A. AND KREITMAN, M. 1986. Evolutionary genetics of two sibling species of *Drosophila, D. simulans* and *D. mauritiana. Evolution*, **40**, 673–691.

COYNE, J. A. AND ORR, H. A. 1988. Two rules of speciation. In: Endler, J. and Otte, D. (eds) *Speciation and its Consequences.* Sinauer Assoc., MA, pp. 180–207.

DOBZHANSKY, TH. 1936. Studies on hybrid sterility. II. Localization of sterility factors in *Drosophila pseudoobscura* hybrids. *Genetics*, **21**, 113–135.

EVGENEV, M. B. 1971. The pattern of polytene chromosome conjugation and crossing-over in interspecific hybrids of *Drosophila. Theor. Appl. Genet.*, **41**, 249–254.

GOULIELMOS, G. N. AND ALAHIOTIS, S. N. 1989. Interspecific hybridization between *Drosophila* species group *melanogaster* sibling species: Isozymic patterns and reproductive relationships. *Genome*, **32**, 146–154.

MATHER, K. AND JINKS, J. L. 1971. *Biometrical Genetics.* Chapman and Hall, London.

NAVEIRA, H. AND FONTDEVILA, A. 1986. The evolutionary history of *Drosophila buzzatii.* XII. The genetic basis of sterility in hybrids between *D. buzzatii* and its sibling *D. serido* from Argentina. *Genetics*, **144**, 841–857.

NAVEIRA, H. AND FONTDEVILA, A. 1991a. The evolutionary history of *Drosophila buzzatii.* XXII. Chromosomal and genic sterility in male hybrids of *D. buzzatii* and *D. koepferae. Heredity*, **66**, 233–239.

NAVEIRA, H. AND FONTDEVILA, A. 1991b. The evolutionary history of *Drosophila buzzatii.* XXI. Cumulative action of multiple sterility factors on spermatogenesis in hybrids of *D. buzzatii* and *D. koepferae. Heredity*, **67**, 57–72.

ORR, H. A. 1987. Genetics of male and female sterility in hybrids of *Drosophila pseudoobscura* and *D. persimilis. Genetics*, **116**, 555–563.

ORR, H. A. 1989a. Genetics of sterility in hybrids between two subspecies of *Drosophila. Evolution*, **43**, 180–189.

ORR, H. A. 1989b. Localization of genes causing postzygotic isolation in two hybridizations involving *Drosophila pseudoobscura. Heredity*, **63**, 231–237.

ORR, H. A. AND COYNE, J. A. 1989. The genetics of postzygotic isolation in the *Drosophila virilis* group. *Genetics*, **121**, 527–537.

PONTECORVO, G. 1943. Hybrid sterility in artificially produced recombinants between *Drosophila melanogaster* and *D. simulans. Proc. R. Soc. Edinb. Sect. B.*, **61**, 385–397.

RICE, W. R. 1989. Analyzing tables of statistical tests. *Evolution*, **43**, 223–225.

SHRIMPTON, A. E. AND ROBERTSON, A. 1988a. The isolation of polygenic factors controlling bristle score in *Drosophila melanogaster.* I. Allocation of third chromosome sternopleural bristle effects to chromosome sections. *Genetics*, **118**, 437–443.

SHRIMPTON, A. E. AND ROBERTSON, A. 1988b. The isolation of polygenic factors controlling bristle score in *Drosophila melanogaster.* II. Distribution of third chromosome bristle effects within chromosome sections. *Genetics*, **118**, 445–459.

VIGNEAULT, G. AND ZOUROS, E. 1986. The genetics of asymmetrical male sterility in *Drosophila mojavensis* and *Drosophila arizonensis* hybrids: Interactions between the Y-chromosome and autosomes. *Evolution*, **40**, 1160–1170.

ZOUROS, E., LOFDHAL, K. AND MARTIN, P. A. 1988. Male hybrid sterility in *Drosophila*: Interactions between autosomes and sex chromosomes in crosses of *D. mojavensis* and *D. arizonensis. Evolution*, **42**, 1321–1331.