Genet. Res. Camb. (1996), **67**, *pp.* 27–41

Recombination load associated with selection for increased recombination

B. CHARLESWORTH[1]* AND N. H. BARTON[2]

[1] *Department of Ecology and Evolution, The University of Chicago, 1101 E. 57th St., Chicago, IL 60637-1573, U.S.A.*

[2] *Institute of Cell, Animal and Population Biology, University of Edinburgh, King's Buildings, West Mains Road, Edinburgh EH9 3JT, UK*

(*Received 17 January 1995 and in revised form 5 July 1995*)

Summary

Experiments on *Drosophila* suggest that genetic recombination may result in lowered fitness of progeny (a 'recombination load'). This has been interpreted as evidence either for a direct effect of recombination on fitness, or for the maintenance of linkage disequilibria by epistatic selection. Here we show that such a recombination load is to be expected even if selection favours increased genetic recombination. This is because of the fact that, although a modifier may suffer an immediate loss of fitness if it increases recombination, it eventually becomes associated with a higher additive genetic variance in fitness, which allows a faster response to direction selection. This argument applies to mutation-selection balance with synergistic epistasis, directional selection on quantitative traits, and ectopic exchange among transposable elements. Further experiments are needed to determine whether the selection against recombination due to the immediate load is outweighed by the increased additive variance in fitness produced by recombination.

1. Introduction

Recombination rates are maintained at high levels in higher organisms, and (in *Drosophila* at least) are highly heritable (Brooks, 1988). Yet the most obvious effect of recombination is to break up combinations of genes that have been built up by selection, thereby reducing mean fitness, and favouring the spread of modifiers that reduce this genetic randomization (Fisher, 1930; Feldman *et al.* 1980; Altenberg & Feldman, 1987; Zhivotovsky *et al.* 1994). This theoretical expectation is apparently supported by several experiments on fitness components in *Drosophila*, which suggest that genetic recombination causes an immediate reduction in fitness. Our purpose in this paper is primarily to show that the existence of a recombination load is to be expected, even when selection maintains non-zero rates of genetic recombination. We also discuss ways in which alternative explanations of recombination load might be tested experimentally.

The first relevant experiments were conducted by Dobzhansky and his collaborators, who showed that the mean egg-to-adult viability of homozygotes for recombinant chromosomes extracted from crosses between chromosomes isolated from nature was lower than the mean homozygous viability of their non-recombinant ancestors (Spassky *et al.* 1958; Spiess, 1958*a*, *b*; Dobzhansky *et al.* 1959; Levene, 1959). Wasserman (1972) found that *D. subobscura* females which were heterozygous for inversions laid eggs with a higher hatchability than homokaryotypic females, but differences in chromosomal constitution and the associated differences in recombination rates were partly confounded with differences in levels of genic heterozygosity (Charlesworth & Charlesworth, 1975).

Mukai & Yamaguchi (1974) found that homozygotes for chromosomes extracted from wild-caught male *D. melanogaster* (*M* chromosomes) had higher viability than homozygotes for chromosomes extracted from females (*F* chromosomes). The ordering of viabilities for chromosomal heterozygotes was $M/F > M/M > F/F$, although the differences were very small. The ordering of genetic components of variance in viability was $M/M < M/F < F/F$. In *Drosophila*, female-derived chromosomes experience one more generation of recombination than male-derived chromosomes, so that the differences between *M* and *F* chromosomes presumably reflect the effects of recombination. Similarly, Charlesworth & Charlesworth (1975) found that the mean viability of heterozygotes

* Corresponding author: B. Charlesworth, Department of Ecology and Evolution, The University of Chicago, 1101 E. 57th St., Chicago, IL 60637-1573, U.S.A. Phone: 312-702-8942; Fax: 312-702-9740; E-mail: bcworth@pondside.uchicago.edu.

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

for *M* second chromosomes of *D. melanogaster* was higher than that of *F/F* homozygotes, and that *M/F* heterozygotes had intermediate viability. While they concluded that this result was not statistically significant (but see the Discussion section of this paper), they observed that females heterozygous for the balancer chromosome *SM5* and *M* chromosomes had a significantly higher fecundity than *SM5/F* females.

Despite a number of ambiguities, which may in part reflect the difficulty of detecting minor differences in fitness components, these experiments suggest that *Drosophila* populations suffer a 'recombination load' (Charlesworth & Charlesworth, 1975), such that a single generation of exposure to recombination produces chromosomes which are somewhat less fit than an equivalent set of chromosomes which have not experienced recombination. The existence of such a recombination load has often been interpreted in terms of the idea that loci in a population at equilibrium between epistatic selection and recombination may exhibit linkage disequilibrium, such that combinations of alleles which are associated with higher fitness are present in excess of their frequency under random combination (Fisher, 1930). Non-recombinant gametes sampled from such a population would be expected to be associated with higher fitness than recombinant gametes, since they are associated with fitter gene combinations (Wasserman, 1972; Mukai & Yamaguchi, 1974; Charlesworth & Charlesworth, 1975). Such an effect provides the driving force for the evolution of reduced recombination in such systems (Fisher, 1930; Maynard Smith, 1978; Feldman *et al.* 1980; Zhivotovsky *et al.*, 1994; Barton, 1995).

An alternative possibility, suggested by Tucíc *et al.* (1981) and Kondrashov (1984), is that recombination is associated with a direct cost in terms of fitness, e.g. because of the induction of deleterious mutations during the chromosome breakage and rejoining events involved in recombination. This seems somewhat unlikely to us, for several reasons. First, meiotic mutants that reduce or eliminate genetic recombination tend to cause a drastic reduction in fitness, partly because of increased frequencies of chromosome non-disjunction due to the production of non-exchange tetrads (Baker *et al.* 1976), and partly because of the connection between DNA repair and recombination (Radman & Wagner, 1993). Less efficient repair would presumably lead to lower fitness. Indeed, the association between DNA repair and recombination has been proposed as a major factor in the evolution of sex and recombination (Bernstein *et al.* 1988). Second, measurements of mutation rates in *Drosophila* indicate that they are similar in males and females, whereas a higher mutation rate would be expected for females if recombination were mutagenic (Woodruff *et al.* 1983). Third, there is no evidence for a difference in the rate of molecular evolution between genes located in regions of the *Drosophila* genome with different rates of recombination, as might be expected if recombination is mutagenic (Begun & Aquadro, 1992).

Both of these interpretations of recombination load imply that there is a fitness cost to recombination, suggesting that natural selection should favour a reduction in the rate of genetic recombination to near zero ('congealing of the genome', Turner, 1967), in the absence of opposing forces. The purpose of this paper is to emphasize a third possibility: that natural selection can actively maintain non-zero rates of recombination, but nevertheless a recombination load can be detected experimentally. (This was previously suggested by Kondrashov (1984) on the basis of a heuristic argument.) This might seem paradoxical, but we shall show that it is a logical consequence of situations in which the advantage to increased recombination accrues from the fact that recombination increases the additive genetic variance in fitness. Under these circumstances, an increase in variance due to increased recombination may lead to a reduction in mean fitness after one generation, because of the production of more extreme, less fit, genotypes. In the long term, however, the greater efficacy of selection with increased additive variance can lead to an increased mean fitness for a population with a higher level of recombination (Kondrashov, 1984; Charlesworth, 1989; Barton, 1995). There may also be selection for modifier alleles which increase genetic recombination, since such modifiers can become associated with lineages with increased additive variance and hence increased fitness. We also show that a similar situation results from the containment of the spread of transposable elements by ectopic exchange, a process proposed by Langley *et al.* (1988).

2. Theoretical analysis

(i) *Mutation-selection balance*

The first situation that we consider is when deleterious mutations are maintained at a large number of loci by mutation, and interact synergistically so that the logarithm of fitness is a concave, decreasing function of the number of mutations carried by an individual. This model has been shown to select for non-zero rates of genetic recombination among the loci concerned (Feldman *et al.* 1980; Kondrashov, 1984; Charlesworth, 1990; Barton, 1995).

If the population is random-mating, as we shall assume here, deleterious mutations are carried mainly as heterozygotes. With equal effects of each locus, fitness is purely a function of the number of heterozygous mutations per individual, *n*. Following Charlesworth (1990), assume that we can represent the natural logarithm of fitness as a quadratic function of *n*:

$$\log(w(n)) = -(\alpha n + \tfrac{1}{2}\beta n^2). \quad (1)$$

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

If the mean number of new deleterious mutations per zygote per generation is U, under some simplifying assumptions the equilibrium mean and variance of the number of mutations per individual, $\bar{n}$ and V_n, and the population mean fitness, $\bar{w}$, can be determined as described by Charlesworth (1990). If chromosomes are drawn from an equilibrium population, and exposed to a change in the frequency of recombination without any selection, only the variance of the distribution of the number of deleterious mutations per individual will be changed. Under synergistic epistasis, increased recombination leads to an increase in V_n, and vice-versa (Kondrashov, 1984; Charlesworth, 1990; Shnol & Kondrashov, 1993; Barton, 1995).

The response of mean fitness to a small change in V_n is measured by the partial derivative of $\bar{w}$ with respect to V_n. Assuming a normal distribution of numbers of mutations per individual, and weak epistasis ($\beta\bar{n} \ll 1$), the results of Charlesworth (1990, p. 216) imply that:

$$\frac{\partial \log(\bar{w})}{\partial V_n} \approx \frac{\beta}{2}\{(2\alpha\bar{n}+\beta\bar{n}^2)-1\}+\frac{\alpha^2}{2}. \quad (2)$$

As pointed out by Charlesworth (1990), if $\bar{n}$ is sufficiently small, the sign of this expression is the same as that of $\alpha^2-\beta$, which is negative except when synergistic epistasis is very weak. The case of $\alpha = 0$ corresponds to multiplicative fitnesses; the case where $\beta = \alpha^2$ is equivalent to the linear fitness model of Kondrashov (1984), for which variance in n does not affect $\bar{w}$. With $\beta > \alpha^2$, a positive value of the derivative is likely only if the mutation rate U for the sector of the genome under consideration is sufficiently high that the term $2\alpha\bar{n}+\beta\bar{n}^2$ becomes significant. For the *Drosophila melanogaster* second chromosome, which constitutes about 40% of the genome, U is probably between 0·2 and 0·5 (Crow & Simmons, 1983; Keightley, 1994). Good data on the values of the selection parameters α and β for net fitness are currently lacking, but Mukai's data on the decline in viability of chromosomes that have accumulated mutations for a long time suggest values of 0·002 and 0·0008 for this component of fitness (Mukai, 1969; Crow, 1970; Charlesworth, 1990). With these parameters, mean fitness is increased by a reduction in variance if $U \leqslant 1$ (Charlesworth, 1990), representing an immediate recombination load.

The magnitude of the expected effect of recombination on mean fitness can be estimated as follows. At equilibrium, the reduction in the linkage disequilibrium component of V_n due to selection must balance the increase due to recombination. In a *Drosophila* population, half the autosomes in a zygote have experienced no crossing over in the parental generation (the paternal chromosomes), and one-half have experienced some crossing over (the maternal chromosomes). The reduction in the variance of the number of deleterious mutations among diploid progeny formed from pairs of independent male-derived chromosomes, when compared with that for the overall population, is thus equal to the effect of selection on the linkage disequilibrium component of the variance. The reduction compared with the variance of female-derived chromosomes is twice this. Noting that approximately half of the effect of selection on variance is associated with the change in linkage disequilibrium, and using the equation of Charlesworth (1990, p. 202) for the net effect of selection on variance, we have

$$V_M - V_F = -\beta V_n^2/(1+\beta V_n), \quad (3)$$

where V_M and V_F are the variances of the numbers of mutations for male- and female-derived chromosomes respectively.

Given equilibrium value of $\bar{n}$ and V_n, eqns (2) and (3) can be combined to yield an approximate expression for the expected mean fitnesses of M/M and F/F flies, $\bar{w}_M$ and $\bar{w}_F$, using the first term of the Taylor series expansion of mean fitness to obtain the effect of a change in variance on mean fitness. The recombination load may be defined as the difference between these, relative to the value for M/M. Similarly, the corresponding relative difference in the genetic variance of fitness (which is approximately the same as the relative difference in the variances of the numbers of mutations) can be found from eqns (2) and (3). For the purposes of illustration, we have used equilibrium values of $\bar{n}$ and V_n, for populations with free recombination, calculated by the approximate method of Charlesworth (1990, p. 205). Numerical studies have shown that chromosomes with a map length similar to that of a *Drosophila* chromosome give nearly the same means and variances as with free recombination, so that there should be no appreciable error in assuming free recombination.

The results for several different assumptions about the selection and mutation parameters are shown in Table 1. It will be seen that, for U values that seem reasonable for a single chromosome, there is always an increase in fitness associated with non-recombinant chromosomes, although this is usually of the order of 10^{-4} or less. The corresponding proportional reduction in variance is larger, often as much as 10^{-2} or more. Of course, since the equilibrium genetic variance in fitness is usually quite small (of the order of 0·004 for a mutation rate of 0·2), the absolute change in variance is of similar magnitude to the recombination load. As noted by Charlesworth (1990), high U values can lead to a reduction in fitness for the non-recombinant chromosomes. In all the cases shown here, non-zero recombination rates are maintained by selection, in the sense that a modifier gene increasing the amount of recombination is selected for in populations with near-zero rates of recombination. For small U values there is usually an ESS value of less than one-half for the average frequency of recombination between pairs of loci (Charlesworth, 1990).

A similar calculation can be performed for the

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

Table 1. *Recombination load with mutation-selection balance and synergistic epistasis. U: genomic mutation rate; $\bar{n}$: mean and variance of the number of deleterious mutations; $\bar{W}_M$, $\bar{W}_F$: mean fitness of M/M and F/F flies, respectively; V_M, V_F: variance of fitness of M/M and F/F flies*

U	$\bar{n}$	V_n	$\frac{(\bar{W}_M-\bar{W}_F)}{\bar{W}_M}$ ($\times 10^{-5}$)	$\frac{(V_M-V_F)}{V_M}$ ($\times 10^{-3}$)	$\bar{n}$	V_n	$\frac{(\bar{W}_M-\bar{W}_F)}{\bar{W}_M}$ ($\times 10^{-5}$)	$\frac{(V_M-V_F)}{V_M}$ ($\times 10^{-3}$)
	$\alpha = 0{\cdot}002, \beta = 0{\cdot}0008$				$\alpha = 0{\cdot}002, \beta = 0{\cdot}002$			
0·1	10·1	10·0	1·39	−3·99	6·68	6·60	3·79	−6·55
0·3	18·4	18·2	3·38	−7·22	12·0	11·8	8·93	−11·6
0·5	24·3	23·8	3·80	−9·43	15·8	15·3	9·95	−15·1
0·7	29·0	28·4	2·59	−10·1	18·9	18·2	6·72	−17·9
0·9	33·2	32·4	−0·29	−12·8	21·6	20·8	−8·22	−20·0
	$\alpha = 0{\cdot}001, \beta = 0{\cdot}002$				$\alpha = 0{\cdot}0005, \beta = 0{\cdot}005$			
0·1	6·92	6·83	4·09	−6·78	4·52	4·42	10·7	−10·9
0·3	12·3	12·0	9·47	−11·9	8·00	7·70	20·4	−18·9
0·5	16·0	15·6	10·1	−15·3	10·4	9·96	26·3	−24·3
0·7	19·1	18·4	7·53	−18·1	12·5	11·8	17·4	−28·7
0·9	21·8	21·0	0·09	−20·1	14·2	13·4	−2·90	−32·4

truncation selection model of synergistic epistasis, discussed by Kondrashov (1984, 1995). We assume as before that the number of mutations per individual at birth is normally distributed, despite the departures from normality caused by truncation selection for the post-selection population. Numerical studies have shown that these departures are usually unimportant (Kondrashov, 1995). If only individuals for whom n is less than the truncation point T can survive, the mean fitness of the population is given by the cumulative probability of the standardized normal variate $z = (T-\bar{n})/\sqrt{V_n}$. From standard results on the effects of truncation selection (Bulmer, 1985, p. 153), the change in mean and variance due to selection are given by

$$\Delta_s \bar{n} = -c\sqrt{V_n} \tag{4a}$$

$$\Delta_s V_n = -c(c+z)\, V_n, \tag{4b}$$

where c is the ratio of the ordinate of the standardized normal distribution at z to $\bar{w}$.

Equating the net change in $\bar{n}$ due to selection and mutation to zero, and noting that with free recombination $\Delta_s V_n = V_n - \bar{n}$ at equilibrium (Charlesworth, 1990, p. 204), we obtain the equilibrium expressions

$$V_n \approx (U/c)^2 \tag{5a}$$

$$\bar{n} \approx V_n(1 + c[c+z]). \tag{5b}$$

Using the same argument that led to eqn (3), we also have

$$V_M - V_F \approx -c(c+z)\, V_n. \tag{5c}$$

The mean fitness of the non-recombinant M chromosomes is given by the cumulative probability of the standardised normal deviate $z' = \sqrt{(V_n/V_M)}$.

Calculations using these results indicate that substantially larger effects on both the mean and variance of fitness as a result of suppression of recombination can be obtained than with the previous fitness model. For example, for $U = 0{\cdot}2$ and truncation selection with 97·5% of the population surviving, we have $\bar{n} = 12{\cdot}05$ and $V_n = 10{\cdot}72$. The relative change in variance due to suppressed recombination is approximately $-0{\cdot}12$, and the relative change in $\bar{w}$ is $7{\cdot}0 \times 10^{-3}$. In contrast to the case of quadratic exponential selection, it is not known whether or not a modifier increasing recombination away from zero is always selected for under truncation selection (Kondrashov, 1984). But Felsenstein (1974) showed that the difference in mean fitness between equilibrium populations with and without recombination determines whether a completely recessive modifier that increases recombination will invade a non-recombining population. Kondrashov's (1984) numerical results demonstrate that truncation selection against deleterious mutations generates such a difference in favour of a recombining population. This implies that a recessive modifier that increases recombination will always be favoured against zero recombination. The greater recombination load associated with truncation selection compared with synergistic epistasis presumably reflects the larger contribution of epistasis compared with additive effects, to the variance in fitness with truncation selection (see Section [iii]).

(ii) *Directional selection on a metrical trait*

In this section, we consider a model of nor-optimal selection on an additively inherited quantitative trait z, such that the optimum value for the trait changes by

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

Table 2. *Recombination load with directional selection. For definitions, see caption to Table 1.* $\Delta\theta$ *is the change in optimum per generation*

$\Delta\theta$	$\frac{(\bar{W}_M-\bar{W}_F)}{\bar{W}_M}$ ($\times10^{-4}$)	$\frac{(V_M-V_F)}{V_M}$ ($\times10^{-4}$)	$\frac{(\bar{W}_M-\bar{W}_F)}{\bar{W}_M}$ ($\times10^{-4}$)	$\frac{(V_M-V_F)}{V_M}$ ($\times10^{-4}$)	$\frac{(\bar{W}_M-\bar{W}_F)}{\bar{W}_M}$ ($\times10^{-4}$)	$\frac{(V_M-V_F)}{V_M}$ ($\times10^{-4}$)
$V_S=20$, $V_E=1$, $V_G=0{\cdot}4$				$V_G=1{\cdot}0$		$V_G=2{\cdot}0$
0	0·98	−0·11*	5·95	−0·89*	22·7	−4·39*
0·02	0·98	−0·10	5·95	−0·88	22·7	−4·37*
0·04	0·98	−0·07	5·94	−0·83	22·7	−4·31*
0·06	0·97	−0·04	5·93	−0·76	22·6	−4·21*
0·08	0·96	−0·01	5·91	−0·67	22·6	−4·07*
0·10	0·96	−0·01	5·89	−0·57	22·6	−3·90
$V_S=10$, $V_E=1$, $V_G=0{\cdot}4$				$V_G=1{\cdot}0$		$V_G=2{\cdot}0$
0	3·84	−0·74*	22·7	−5·19*	82·6	−21·8*
0·02	3·84	−0·70	22·7	−5·14*	82·6	−21·7*
0·04	3·83	−0·59	22·6	−5·04*	82·6	−21·5*
0·06	3·81	−0·45	22·6	−4·78*	82·5	−21·3*
0·08	3·79	−0·28	22·5	−4·47	82·4	−20·9*
0·10	3·73	−0·07	22·5	−4·10	82·3	−20·4*

* These cases fail to satisfy the condition for non-zero recombination to be favoured by selection.

an amount $\Delta\theta$ per generation (Charlesworth, 1993; Barton, 1995). Let the sum of the environmental variance V_E and the intensity of stabilizing selection ω^2 be V_S; the additive genetic variance in the trait is V_G. Charlesworth (1993, eqn (5)) implies that the response of population mean fitness to a change in variance (holding the mean constant) is given approximately by

$$\frac{\partial \log(\bar{w})}{\partial V_G} \approx \frac{V_S(\phi-(V_G/V_S)^2)}{2V_G^2}, \qquad (6)$$

where $\phi = (\Delta\theta)^2/V_S$ (it is assumed that selection is sufficiently weak that $V_G \ll V_S$).

It follows that the sign of the effect of an increase in variance on mean fitness is the same as that of the term in braces. Charlesworth (1993, eqns (9)) implies that the condition for non-zero recombination to be favoured by selection is much less stringent than the condition for increased variance to result in increased fitness (the condition for selection for non-zero recombination is approximately $2\phi > (V_G/V_S)^3$). This model will therefore often produce a recombination load under conditions when non-zero recombination is maintained by selection.

The magnitude of the effect of suppressing recombination on mean fitness can be predicted in a similar way to that used for the mutation selection balance model. In this case, the change in variance of the quantitative trait due to selection over one generation is $\Delta_s V_G \approx -V_G^2/V_S$, which can be equated to twice the change in variance due to linkage disequilibrium (Bulmer, 1985, p. 159), and hence to the difference in variance between M and F chromosomes in *Drosophila*. The proportional increase in mean fitness for non-recombinant chromosomes is obtained by multiplying eqn (6) by the change in variance. The change in the variance of fitness due to recombination suppression can similarly be calculated from the derivative of the genetic variance in fitness. This variance can be obtained as follows. Integrating over the distribution of phenotypic values associated with a given genetic value z_g (assumed to be normal, with variance V_E) gives the mean fitness associated with z_g as:

$$w(z_g) = \frac{\omega}{\sqrt{V_s}}\exp\left(-\frac{(z_g-\theta)^2}{2V_s}\right).$$

The variance of fitness is obtained by further integration of the squared deviations of $w(z_g)$ from the mean fitness over the distribution of z_g (assumed to be normal with variance V_G). This yields the following expression for the genetic variance of fitness, V_{Gw}

$$V_{Gw} = \frac{\omega^2}{\sqrt{[V_s(2V_G+V_s)]}}\exp\left(-\frac{(z_g-\theta)^2}{(2V_G+\omega^2)}\right)-\bar{w}^2, \qquad (7a)$$

where

$$\bar{w} = \frac{\omega}{\sqrt{(V_G+V_s)}}\exp\left(-\frac{(z_g-\theta)^2}{2(V_G+V_s)}\right)$$

and $\quad V_S = V_E+\omega^2 \qquad (7b)$

Note that eqn 7*a* gives the variance of individual fitness, rather than of gametic fitness, and therefore includes dominance components (see Section iv).

Some representative results are shown in Table 2. The value of 20 for V_S has been suggested to be

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

representative of the intensity of selection on quantitative traits by Turelli (1984); the value of 10 represents rather intense selection. The cases where $\Delta\theta = 0$ represent populations that are in equilibrium under pure stabilizing selection. From Fisher's Fundamental Theorem, this corresponds to a situation when all the variance in fitness is non-additive in nature, and so produces a selection pressure for reduced recombination through epistatic selection (Charlesworth, 1993). As $\Delta\theta$ increases, the advantage to increased recombination from breaking down negative linkage disequilibria among the loci controlling the trait becomes greater. But if the heritability of the trait is high, this advantage is weaker, and may never overcome the disadvantage from epistatic selection (see right-hand part of Table 2).

It can be seen that there is always an immediate fitness advantage to suppressed recombination, associated with a decrease in the variance in fitness, even when non-zero recombination is favoured by selection. The magnitude of the recombination load only depends weakly on $\Delta\theta$ over the range of parameters shown here, which are chosen so that the load due to directional selection is moderate (Charlesworth, 1993). A recombination load of at most $\approx 10^{-3}$ is compatible with the existence of selection in favour of non-zero recombination under these conditions; this is considerably larger than in the case of mutation-selection balance with quadratic epistasis. As with mutation and selection, the proportional reduction in variance due to recombination suppression is much larger than the recombination load. Much larger recombination loads can arise with parameter values for which there is a selective disadvantage of non-zero recombination. It should be noted that we have assumed in these calculations that recombination is suppressed throughout the whole genome; the effect of suppressing recombination on a chromosome that contributes a fraction p of the genome means that the tabulated values should be multiplied by p.

(iii) *The general relation between variation in fitness and selection on recombination*

In both the above examples, recombination is subject to two opposing pressures. Modifiers that increase recombination tend to decrease in frequency because they produce unfavourable gene combinations. They tend to increase because they become associated with an increased additive variance in fitness, and hence with a more effective response to directional selection. When epistasis is weak, the rate of change of the modifier due to these two pressures can be expressed in terms of the variance in log (fitness), which is in turn related to the recombination load (Barton, 1995). This general approximation is possible because, with weak epistasis, linkage disequilibria rapidly settle into a 'quasi-equilibrium' between selection and recombination (Barton & Turelli, 1991). Here, we summarize the argument, and discuss how it can be used to measure the selection on recombination. Details are given in the Appendix.

Selection is assumed to be weak relative to recombination, so that linkage disequilibria between particular sets of loci are small, and change slowly relative to the timescale of recombination. We make the further assumption that the directional selection acting on each locus is strong relative to the epistasis between sets of two or more loci; the relevant coefficients are assumed to be $O(s)$ and $O(s^2)$ respectively. (Here, s is a measure of the strength of directional selection on each locus.) If directional selection is in fact of the same order or weaker than epistasis (as would be the case for a population at equilibrium under epistatic selection and recombination), then the results simplify, to show that recombination is selected against (Zhivotovsky *et al.* 1994; Barton, 1995).

Throughout, we work with the average effect of a gamete on the fitness of individuals formed by combining it with gametes sampled randomly from the population (see Appendix), which we denote by W^* to distinguish it from the fitness of the diploid genotype, W. W^* does not include any contribution from dominance interactions, and may be regarded as a measure of gametic fitness. To leading order, the variance of $\log(W^*)$ is:

$$\mathrm{var}(\log(W^*)) = V_1 + v_1 + \sum_{k>1} V_k + O(s^5). \qquad (8)$$

(This is equivalent to Barton, 1995, eqn (13*b*).)

These variance components are defined in terms of the selection coefficients (see Appendix). We use the distribution of $\log(W^*)$, because the components of variance of $\log(W^*)$ reflect the deviations from *multiplicative* fitness which generate linkage disequilibria (Feldman *et al.* 1980; Shnol & Kondrashov, 1993). In eqn 8, $V_1 + v_1$ is the additive variance in $\log(W)$, whilst the V_k are the components of non-additive variance due to sets of k interacting loci; these are defined in terms of selection coefficients in eqn A 3*b*. V_1 is the genic contribution to the additive variance in $\log(W^*)$ i.e. the variance that is contributed by the sum of the variances at each locus. Linkage disequilibria alter the additive genetic variance, making a contribution denoted by v_1. Given the approximation that epistasis is weak, the non-additive variance component V_k is dominated by genic contributions due to heterozygosities at the loci involved; it is not significantly influenced by linkage disequilibrium (i.e. $v_k \ll V_k, v_1$). (Note that Barton (1995) denotes components of $\log(W^*)$ by V'_k, rather than by V_k; here, we drop the primes.)

We now relate these variance components to the selection on a modifier, and to the recombination load. Suppose the modifier allele is at frequency p, and increases recombination between loci j and k from r_{jk} to $r_{jk} + \delta r_{jk}$ ($\delta r_{jk} \ll r_{jk}$). For simplicity, we consider an

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

unlinked modifier. Then, from Barton (1995, eqn 14), the selection coefficient on the modifier is:

$$\frac{\Delta p}{pq} \approx -\left(\frac{3v_1}{2}+2V_2\right)E\left[\frac{\delta r_{jk}}{(1+r_{jk})}\right]-\sum_{|N|>2} V_{|N|}E\left[\frac{\delta r_N}{(1+r_N)}\right] \qquad (\cdot)$$

The first term gives the effect of pairwise interactions. The second term is a sum over all higher-order interactions; r_N is the rate at which recombination breaks up a set N of loci, and δr_N is the effect of the modifier on this rate. (For example, r_{jkl} is the total rate of all recombinations which break up the three loci $\{j,k,l\}$.) $E[\delta r_N/(1+r_N)]$ represents an expectation over all sets of loci with $|N|$ members, weighted by the associated variance component $V_{|N|}$. Since, with loose linkage, large sets are almost certain to recombine ($r_N = 1$), δr_N may be small, and these higher-order terms negligible. The fate of the modifier is then determined by the factor $((3/2)v_1+2V_2)$, and is independent of the linkage relations among the selected loci. If linkage disequilibria reduce the additive genetic variance in log(fitness) by more than four-thirds of the nonadditive variance ($v_1 < 0$, $-(4/3)V_2$), and if higher-order epistasis ($V_3, V_4 \ldots$) is negligible, then recombination will increase. The approximation also applies to linked modifiers. In that case recombination is more likely to be favoured, because modifiers remain associated with favourable genes for longer (Barton, 1995, eqn 14).

Both the contribution of linkage disequilibria to the additive variance in $\log(W^*)$, v_1, and the non-additive variance V_2 can (at least in principle) be measured. Suppose that a sample of chromosomes is extracted from a population, each being held against a balancer chromosome. The fitness W^* of each chromosome can then be measured using heterozygotes between that chromosome and some set of standard chromosomes chosen from the same sample. (In practice, some component of fitness such as larval viability would be measured, under conditions as close as possible to the original environment.) Suppose that the chromosomes were sampled immediately after selection, without recombination. If (as is necessary for recombination to be favoured) epistasis is negative ($v_1 < 0$) then the variance of $\log(W^*)$ would have been reduced below v_1 by selection, by $v_1 E[r_{jk}]$. One round of recombination would return $\mathrm{var}(\log(W^*))$ to the value defined for the zygote population, increasing it by $-v_1 E[r_{jk}]$. Successive rounds of recombination under relaxed selection would cause a further net change of $-v_1$ as all linkage disequilibria were dissipated. If (as in the *Drosophila* experiments discussed above) the comparison is between the variance of chromosomes extracted from males (with no recombination) and females (with twice the average value), then the difference should be $2v_1 E[r_{jk}]$. The factor $E[r_{jk}]$ can be estimated from knowledge of the distribution of crossing over along the chromosome (as, for example, in Charlesworth, 1990, eqn 14). Thus, in principle v_1 could be estimated experimentally.

The difference in mean $\log(W^*)$ caused by one round of recombination is equal to the non-additive variance $\Sigma_{k\geqslant 1} V_k$ (see Barton, 1995, eqn 15 *et seq.*). This is because the increase in mean W^* within a generation caused by selection alone is equal to the total variance of $\log(W^*)$, whereas the increase due to allele frequency change in a quasi-equilibrium population is the additive component of the variance. As found for the variance in log fitness, the difference in mean between chromosomes extracted from male and female *Drosophila* would be twice the non-additive variance.

If a population is at equilibrium under epistatic selection and recombination, coefficients of epistasis and directional selection are of the same order. There will be no additive genetic variance ($V_1+v_1 = 0$), the non-additive variance $\Sigma_{k\geqslant 1} V_k$ will be much larger than v_1, the recombination load will be much greater than the increase in additive variance released by recombination, and recombination will be selected against (see eqn A 8*b*). The theory makes no prediction as to whether the total genetic variance will be increased or decreased by recombination, but it does predict that the magnitude of the change in variance will be a fraction of the total genetic variance and of the recombination load, whose size depends on the strength of selection acting in the system and the rates of recombination (eqn A 8). These predictions could in principle be used to find whether epistasis, and the consequent recombination load, are strong enough for recombination to be selected against.

In order for recombination to be favoured by an interaction between directional selection and epistasis, coefficients of pairwise epistasis must be of the same order as the square of the coefficients of direction selection. If this is so, then the additive genetic variance contributed by linkage disequilibria, v_1, and the non-additive variance, V_2, will be comparable, regardless of the number of loci involved and their average allele frequencies (see eqn A 3*b*). Correspondingly, the change in variance in fitness due to recombination must be comparable to the change in mean. The theory also predicts that v_1 and V_2 are small relative to V_1, the genic component of the additive variance. V_1 is $O(s^2)$, whereas v_1 and V_2 are $O(s^4)$. Moreover, V_1 is proportional to the sum of heterozygosities (Σpq) across loci, whereas v_1 and V_2 are proportional to the sum of squares of heterozygosities ($\Sigma(pq)^2$; from eqn A 3*b*); they will therefore make up a small proportion of the genetic variance, especially if variation is due to rare alleles. Therefore, recombination cannot be strongly favoured by an interaction between directional selection and epistasis unless the total heritable variance in fitness is high. This is borne out by the numerical results, for example in Table 2.

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

Table 3. *Components of variance of log(fitness) under mutation/selection balance with free recombination. V_1 is the genic contribution to the additive genetic variance of log(fitness); v_1 is the contribution of linkage disequilibria to this additive variance; and V_2 is the non-additive variance in log(fitness) due to pairwise epistasis. Δp is a change in frequency of the modifier per generation*

U	$\bar{n}$	V_n	V_1	v_1	V_2	$\frac{\Delta p}{pq\delta r}$	
α=0·002, β=0·0008							
0·1	10·05	9·97	0·00101	−0·00001	0·00003	−0·00003	
0·3	18·30	18·03	0·00507	−0·00007	0·00011	−0·00008	
0·5	24·03	23·57	0·01083	−0·00021	0·00019	−0·00004	
0·7	28·70	28·04	0·01788	−0·00041	0·00026	+0·00006	
0·9	32·77	31·91	0·02609	−0·00068	0·00034	+0·00023	

In principle, fluctuations in epistasis can also maintain recombination, an effect which would be hard to investigate in laboratory experiments. However, fluctuating epistasis can only favour recombination under very restrictive conditions (Maynard Smith, 1978; Nee, 1989; Barton, 1995); models of fluctuating *selection* may favour recombination through the effects of directional selection, as discussed here, rather than through fluctuating *epistasis per se* (Charlesworth, 1976; Hamilton, 19880; Maynard Smith, 1980, 1988; Hamilton, 1993).

(iv) *Application of the general theory to mutation/selection balance*

We now illustrate these general arguments by applying them to the specific example of mutation/selection balance. The reasoning is set out in more detail in Barton (1995, Appendix 3), where it is assumed that selection is weak, so that the population reaches a 'quasi-equilibrium'. For plausible parameter values, this is a good approximation (e.g. Barton, 1995, Table 3 and Table 3 below). We will use these to derive the components of the variance in log(fitness), and show how these relate to the recombination load and to selection on modifiers of recombination.

First, consider a mutation/selection balance, with fitness given by the Gaussian function of eqn 1 above. Since mutations are assumed to be very rare, only heterozygous effects need be considered. Thus, dominance components can be neglected, and the individual fitness of a genotype, W, is equal to the average of the fitnesses, W^*, contributed by the two gametes. The mean and variance of log(fitness) can thus be derived by taking expectations over eqn 1, and are given in terms of the moments of the number of deleterious mutations. Assuming that this number is normally distributed:

$$\overline{\log(W^*)} = -\alpha\bar{n} - \frac{\beta}{2}\bar{n}^2 - \frac{\beta}{2}V_n \qquad (10a)$$

$$\mathrm{var}(\log(W^*)) = -(\alpha+\beta\bar{n})^2 V_n + \frac{\beta^2 V_n^2}{2}. \qquad (10b)$$

Note that the gradient of $\overline{\log(W^*)}$ with respect to the variance of n is $-\beta/2$, a simpler form than eqn 2, which gives the gradient of $\log(\bar{W}^*)$; the mean of the log(fitness) differs appreciably from the log of the mean fitness. The components of variance in log (fitness) can be deduced from these, using eqn A 3*b*. The genic component of the additive genetic variance is $V_1 = (\alpha+\beta\bar{n})^2\bar{n}$, whilst the contribution of linkage disequilibria is $v_1 = (\alpha+\beta\bar{n})^2(V_n-\bar{n})$; together, these sum to give the first term in eqn 10*b*, which is the additive genetic variance. The non-additive variance is entirely due to pairwise interactions, and is given by the last term in eqn 10*b*. This is dominated by the genic contribution ($V_2 = \beta^2\bar{n}^2/2$), linkage disequilibria being negligible ($\bar{n} \approx V_n$).

Under the assumption that the population is in quasi-linkage equilibrium, the variance in the numbers of deleterious mutations is reduced by $V_n - \bar{n} = -\beta\bar{n}^2$ (from Barton, 1995, eqn A 3.5, assuming free recombination). When mutation balances selection, $U = (\alpha+\beta\bar{n})\,V_n = (\alpha+\beta\bar{n})(\bar{n}-\beta\bar{n}^2)$. The mean and variance ($\bar{n}$, V_n) are given by the solution of this cubic equation, and are shown in Table 3. They are close to the more accurate values calculated by Charlesworth (1990; see also Barton, 1995, table 3). Given $\bar{n}$ and V_n, the variance components can be calculated; the selection on an unlinked modifier is then given by $-(v_1+4V_2/3)$. Barton (1995, table 3*b*) shows this is also a good approximation to the results of Charlesworth (1990).

Table 3 gives values for $\alpha = 0{\cdot}002$, $\beta = 0{\cdot}0008$, and $U \leqslant 0{\cdot}9$, a plausible range for *Drosophila*. As expected for weak epistasis, linkage disequilibria make a small contribution to the additive variance, which is comparable with the non-additive variance (v_1, $V_2 \ll V_1$). A reduction in recombination below 50% is favoured for $U < 0{\cdot}6$ (Table 3). For these parameters, modifiers which increase recombination from zero are always favoured (Charlesworth, 1990), implying that there is an intermediate ESS recombination rate for $U < 0{\cdot}6$. The variance components are related to the immediate effect of recombination on the mean and variance of log(fitness). In the previous section, it was argued that the difference in variance of log(fitness)

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

between chromosomes extracted from male and female *Drosophila* is $2v_1 E[r_{jk}]$; if the map length of the chromosome is such that $r_{jk} \approx 1/2$ in females, then $E[r_{jk}] \approx 1/4$ averaged over the two sexes, and so the difference in variance is just $v_1/2$. Similarly, the difference in mean log (fitness) between chromosomes from males and females is twice the non-additive variance, $2V_2$. Note that these values differ from those given in Table 1, because they refer to the distribution of log (fitness) rather than of fitness, and because the difference in variance was there expressed relative to the genetic variance ($(V_M - V_F)/V_M$).

(v) *Ectopic exchange and transposable elements*

The last case which we shall consider is that of transposable elements (TEs) maintained by a balance between transpositional increase in copy number and selection against deleterious chromosome rearrangements induced by ectopic exchange (Langley *et al.* 1988). There is direct experimental evidence for this process in *Drosophila* (Langley *et al.* 1988; Montgomery *et al.* 1991), and many features of the distribution of TEs in *Drosophila* populations can be explained by its role in copy number regulation (Charlesworth *et al.* 1992). Following the notation of Langley *et al.*, let the probability that an ectopic exchange event takes place at meiosis between two genomic regions j and k with copy numbers n_j and n_k, and generates a dominant lethal rearrangement, be $\kappa_{jk} n_j n_k$.

In order to facilitate further calculations, we shall assume uniformity of recombination rates over the genome, so that κ_{jk} takes the same value κ for all j and k. It is then reasonable to assume that the proportion of elements located in a given section of the genome is the same as the proportion of the genome represented by the section in question. Under this assumption, Langley *et al.* (1988, eqns (3), (10) and (13)) imply that the fitness of an individual carrying n elements is approximately $1 - \kappa n^2$. Since copy number in *Drosophila* follows the Poisson distribution to a good approximation (Charlesworth & Langley, 1989), if $\bar{n} \gg 1$ we have

$$L \approx \kappa \bar{n}^2 \qquad (11a)$$

or

$$L \approx \bar{n}u/2, \qquad (11b)$$

since under these assumptions $\bar{n} = u/(2\kappa)$.

Equation (11*b*) gives a value for L that is one-half that predicted by the general expression for load due to transposable elements, given by Charlesworth (1985, eqn (8)). This is because that equation implicity ignores the effects of departure from independence among elements, caused by the synergism in fitness effects generated by the quadratic dependence of the frequency of ectopic exchange events on element abundance. As in the case of mutation-selection balance, such synergism results in an approximate halving of the load (Kimura & Maruyama, 1966; Crow, 1970; Charlesworth, 1990).

If exchange takes place randomly between pairs of elements, independently of their location in the genome (Langley *et al.* 1988, model 1), and if only exchanges between elements located in the segment covered by the balancer are suppressed, a proportion p^2 of the normal frequency of lethal events will be suppressed, and hence there will be a gain in fitness of $p^2\bar{n}u/2$ due to recombination suppression. If exchange takes place only between elements located in the same small region of the chromosome (Langley *et al.* 1988, model 2), the relevant proportion of exchanges affected is p and the associated fitness gain is $p\bar{n}u/2$. The recombination load in terms of the difference between M and F chromosomes is twice this. There is empirical evidence which provides some support for model 2 for *Drosophila* (Davis *et al.* 1987), so that the larger of these two values may be more relevant.

A recent estimate of the net rate of movement of TEs in *D. melanogaster* suggests a value of $\bar{n}u$ of about 0·10 for 17 of the 40 or so element families (Nuzhdin & Mackay, 1995). This may be a fairly good estimate for the whole genome, since the observed transposition events all involve four unusually mobile element families. Highly mobile elements are more likely to be picked up in cloning studies, so that the failure to include a number of less mobile elements in the study will probably have only a small overall effect on the estimated genomic rate of movement. Given that a second chromosome balancer in *D. melanogaster* comprises about 40% of the genome, the estimated $\bar{n}u$ value predicts a recombination load for this chromosome due to ectopic exchange of between 0·016 and 0·044.

This seems a surprisingly high value, but the conclusion should be qualified in three respects. First, some ectopic exchange events will generate detrimental rather than lethal rearrangements (e.g. duplications or small deletions, Montgomery *et al.* 1991). These will persist in the population for several generations while contributing to the overall load. If the mean persistence time of a class of rearrangement is t, its contribution to the recombination load observed over just one generation is weighted by $1/t$. If ectopic exchange generates a large fraction of detrimental rather than lethal rearrangements, the load could be much less than the above estimate. Second, the experimental procedures used for measuring recombination load in *Drosophila* usually involve the storage of extracted chromosomes (without exposure to recombination) for several generations before assay of their fitness effects. This means that dominant lethal or highly deleterious chromosomes will not be included, and so a large fraction of the load associated with ectopic exchange will not be detected. Third, population studies show clearly that forces other than meiotic ectopic exchange (such as selection against

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

insertional mutations or against rearrangements induced by mitotic ectopic exchange) contribute to the regulation of TE abundance (Charlesworth *et al.* 1992), so that only a portion of the load associated with TEs is attributable to ectopic exchange. Observed associations between restricted meiotic recombination and the abundance of TEs suggest, however, that meiotic ectopic exchange must play a major role (Charlesworth *et al.* 1992; Sniegowski & Charlesworth, 1994).

The fitness variance associated with ectopic exchange can be determined as follows, again assuming uniformity of the frequency of exchange in different regions of the genome, and a Poisson distribution of copy number. The fitness variance associated with variance in copy number is then

$$V_w \approx 4L^2/\bar{n}. \tag{12}$$

Since $\bar{n}$ for the total of all *D. melanogaster* elements is over 100, eqn (12) implies that the fitness variance contributed by ectopic exchange is a small fraction of the total fitness variance of at least 0·01 from all sources (Charlesworth, 1987). In addition, for the reasons given above for the case of load, it is unlikely that a reduction in this variance due to suppression of recombination could be detected experimentally.

Finally, we note that meiotic ectopic exchange helps to create a selection pressure in favour of non-zero rates of recombination. There are two reasons for this. One is the same as for deleterious mutations of the conventional kind; the non-linearity of the relationship between fitness and copy number implies that selection induces negative covariances between the contributions of different loci to copy number, which are diminished by recombination. We shall not consider this process further, since it is essentially the same as the mutation-selection balance model (we have similarly ignored this aspect of ectopic exchange in considering recombination load, to which it is likely to make only a small contribution). The second reason postulates that increased recombination leads to a higher frequency of meiotic ectopic exchange, so that a modifier allele associated with high recombination will eventually come to acquire a lower load of deleterious rearrangements, and hence will experience higher fitness.

This can be analysed in a similar way to that used previously for deleterious mutations (Charlesworth, 1990). Consider a modifier allele that increases the rate of ectopic exchange in its heterozygous carriers by $\delta\kappa$, and which is introduced at low frequency into a population with a zero or near-zero rate of genetic recombination. Using Langley *et al.* (1988, eqns (2) and (4)) for the case of a uniform rate of ectopic exchange, it is easily seen that the presence of the modifier perturbs the per generation change in copy number by $-2\bar{n}\,\delta\kappa$. With the assumed low rate of genetic recombination, Charlesworth (1990, eqn (A 12)) means that the deviation of copy number from the population mean for carriers of the modifier equilibrates at

$$\delta\bar{n} \approx -2\bar{n}\,\delta\kappa \tag{13a}$$

and the corresponding difference in fitness between carriers of the modifier and the rest of the population is

$$\delta\bar{w} \approx 4L\,\delta\kappa. \tag{13b}$$

This shows that a modifier which increases the rate of ectopic exchange will spread in a population with a low frequency of recombination. If meiotic recombination and ectopic exchange are both affected by the modifier, this implies that non-zero recombination is favoured by selection on the rate of ectopic exchange.

The case of a rare dominant suppressor of recombination (such as an inversion), with an associated reduction in the rate of ectopic exchange, can be analysed similarly. It is easily seen that such a recombination suppressor will equilibrate with a higher copy number than in the population at large (Eanes *et al.* 1992). However, unless ectopic exchange is the only force responsible for the elimination of TEs, the equilibrium abundance of TEs associated with the suppressor will be less than inversely proportional to the rate of ectopic exchange in heterozygotes for the suppressor (see Langley *et al.* 1988, eqn (6)). Since the load for the suppressor genotypes is proportional to the product of their frequency of ectopic exchange and the copy number associated with the suppressor, there will at most be no effect of the suppressor on load, and usually a decrease. Thus, ectopic exchange does not protect a freely recombining population from invasion by a dominant recombination suppressor. This indicates the existence of an ESS with intermediate recombination frequencies, as in the other models which we have considered.

3. Discussion

The results presented above establish that the existence of recombination load (Charlesworth & Charlesworth, 1975) is consistent with the operation of forces that promote the maintenance of genetic recombination, as proposed by Kondrashov (1984) for the case of mutation and truncation selection. An intuitive interpretation of this apparently surprising result was given in the Introduction. The observation of recombination load does not, therefore, necessarily imply that selection favours 'congealing of the genome' (Turner, 1967), either because of epistatic variance in fitness maintained by selection (Fisher, 1930; Maynard Smith, 1978; Feldman *et al.* 1980), or because of direct fitness costs of recombination (Tucíc *et al.* 1961; Kondrashov, 1984). It is important to note that recombination load as defined here simply means that a single generation of recombination reduces the fitness of the progeny of the individuals concerned,

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

relative to the fitness of the progeny of comparable individuals in which recombination has not occurred. This does not imply that, in the long term, the mean fitness of a population decreases with an increasing frequency of genetic recombination. The reverse is true for all the examples considered by us.

There is, of course, a question of whether the data on the effects of recombination on the mean and variance of fitness components can be explained by any of the mechanisms discussed above. A fairly strong conclusion from the models is that there can be selection for increased recombination only if there is a fairly large additive variance in gametic fitness, which in turn implies a substantial net heritable variance in fitness. While the latter condition seems to be met (Charlesworth, 1987; Houle, 1992), published experiments only provide information on individual fitnesses, not gametic fitnesses, and no separation of these variance components has been achieved. In Section 2 (iii) we have outlined possible experimental approaches to estimating the magnitudes of the additive and non-additive components of the variance in gametic fitnesses, v_1 and V_2. The proposal is that selection is likely to favour non-zero recombination if v_1 is negative, and of much larger magnitude than V_2. For unlinked loci, and pairwise epistasis, the selection gradient on recombination is equal to $-(v_1+4V_2/3)$. If V_2 is the only significant component of gametic fitness variance, then epistasis is likely to select for decreased recombination. Experiments to separate these components are feasible in principle, although considerable effort would be required.

The published results also suffer from problems of interpretation. The experiments by Dobzhansky and his colleagues for chromosomal homozygotes in three *Drosophila* species (Spassky *et al.* 1958; Spiess, 1958*a*, *b*; Dobzhansky *et al.* 1959) gave recombination loads for egg-to-adult viability ranging from 8·5 to 25%, with a mean of 14%. Their data on the effect of recombination on variance are hard to analyse in terms of the methods described above. It is not clear how to interpret these data, since the sources of variation in fitness for chromosomal homozygotes may not be identical to those for heterozygotes. Furthermore, the chromosomes used were not a random sample of non-lethals, but were chosen to be in the middle range of viability.

Mukai & Yamaguchi's (1974) results on chromosomal heterozygotes gave an ambiguous result for recombination load, although they found a clear indication that fitness variance is increased by recombination (the proportionate difference in variance between M/M and F/F genotypes in their data is 64%, but the sampling error of this estimate is very high). Their data on chromosomal homozygotes gave a recombination load of 3%. The results should be treated with some caution, however, as the method of extraction which they employed means that M chromosomes may have been liable to hybrid dysgenesis (Berg & Howe, 1989) in the first few generations of the crossing procedure. The M chromosomes may therefore have carried more strongly deleterious mutations than the F chromosomes, perhaps explaining the failure to detect a heterozygous recombination load.

The results of Charlesworth & Charlesworth (1975) on the viability of chromosomal heterozygotes gave a recombination load of 0·7%, but the difference between M/M and F/F was apparently non-significant. We have re-analysed their data taking into account the information on M/F individuals, by determining the probability that the differences in means M/M versus M/F and M/F versus F/F each exceed their observed value on the null hypothesis of no recombination load, assuming that they follow a bivariate normal distribution. If each mean has the same sampling variance (estimated to be $1{\cdot}8\times10^{-5}$ in this case), the correlation between the sampling errors of these two contrasts is $-0{\cdot}5$. Using the data of Charlesworth & Charlesworth (1975), we find that $P = 0{\cdot}016$. Their estimate of the difference in fertility between females carrying M and F chromosomes over the *SM5* balancer (7%) is much larger than the recombination load for viability, but reflects the performance of wild chromosomes on an atypical genetic background.

Overall, these experiments suggest that there is a measurable recombination load in *Drosophila*, and that its magnitude may be as high as a few per cent when homozygous chromosomes are studied. The magnitude of the recombination load for heterozygous chromosomes seems to be much smaller, of the order of 0·7% or less. This is still much larger than seems possible under either the quadratic log fitness mutation-selection balance model, or the directional selection model (Tables 1 and 2). It is just about consistent with the truncation selection model of mutation-selection balance (Section 2 [i]). For the reasons given above, it seems unlikely that these experiments could detect more than a fraction of the load caused by ectopic exchange among transposable elements, but the magnitude of this load is sufficiently large that it could be a major contributor to the observed effects. Further experiments of large size are needed if a reliable estimate of the magnitude of recombination load for chromosomal heterozygotes is to be obtained.

The interpretation of experiments on recombination load therefore depends on being able to separate the effects of ectopic exchange from other factors. It is not easy to imagine experiments in which this can readily be done. Tucíc *et al.* (1981) found an inverse correlation between the amount of recombination with a marker chromosome exhibited by an extracted wild chromosome, and the homozygous effect of the wild chromosome on male and female fertility. These results are consistent with the effects of ectopic exchange, but not with the other processes which we have discussed (we have already given reasons for

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

doubting the interpretation that recombination directly lowers fitness, as proposed by Tucíc *et al.* 1981). A repeat of this type of experiment is desirable. Another informative experiment would be to compare the fitness of chromosomes re-extracted from males and females that were originally homozygous for a copy of the chromosome in question. Since ectopic exchange in *Drosophila* seems to be severely reduced in frequency in chromosomal homozygotes (Montgomery *et al.* 1991), we might expect to find only a small or no difference in fitness between such chromosomes if ectopic exchange, as opposed to a direct effect of recombination on fitness, is involved. Finally, we note that the experiments of Coyne *et al.* (1993) on the effects on egg hatchability of heterozygosity for pericentric inversions in female *D. melanogaster* gave the surprising result that 11 out of 30 third chromosomal inversions conferred higher hatchability of eggs produced by heterozygous females. This is consistent with the possibility that the suppression of recombination by heterozygous inversions reduces the frequency of harmful products of crossover events, although (as noted by Coyne *et al.*), differences in the genetic background could also be involved.

This work was supported by NSF grant DEB 9317683, BBSRC grant GR/H/09928, and by the Darwin Trust of Edinburgh. We are grateful for comments on the manuscript from S. P. Otto and A. S. Kondrashov.

Appendix

The change in mean and variance of log(fitness) due to a change in recombination

We use the notation of Barton & Turelli (1991), in which the relative fitness of a diploid individual is written as a polynomial function of genotype:

$$\frac{W}{\bar{W}} = 1 + \sum_U a_{U,\varnothing}(\zeta_U - C_U) + \sum_V a_{\varnothing,V}(\zeta_V^* - C_V) + \sum_{U,V} a_{U,V}(\zeta_U - C_U)(\zeta_V^* - C_V). \quad \text{(A 1)}$$

We assume two alleles at each locus. The state of locus i is labelled as $X_i = 0$ or 1, allele frequencies are $p_i = E(X_i)$, $q_i = 1 - p_i$, and $\zeta_i = (X_i - p_i)$ denotes the deviation of locus i from the mean. U, V denote sets of loci (e.g. $U = \{i,j,k\}$), $\varnothing$ denotes the empty set $\{\}$, and ζ_U denotes the product of the ζ_i over the set U. Linkage disequilibria are defined by the central moments, $C_U = E(\zeta_U)$. Allele frequencies and linkage disequilibria are defined immediately after random union of gametes, so that $C_{U,V} = C_U C_V$. The selection coefficients $a_{U,V}$ represent selection on the combination of alleles at loci U from the maternal genome, and V from the paternal genome. We assume selection on diploid viability, with no cis/trans effects, so that $a_{U,\varnothing} = a_{\varnothing,U}$, and $a_{U,V} = (|UV|!/|U|!|V|!)a_{UV,\varnothing}$, where the sets U, V consist of distinct loci. (The coefficient $a_{i,i}$ represents dominance between homologous alleles at locus i, and is not necessarily equal to $2a_{ii,\varnothing}$.)

Mating is random, and selection acts on viability, or through multiplicative effects on fertility. The response to selection and recombination is then approximately independent of dominance, and with weak selection depends only on the marginal fitnesses of the gametes, W^*. These are defined by considering zygotes formed by taking one intact haploid genome from the genotype in question, and pairing that genome with another, taken at random from the population. By analogy with the breeding value, $(W^* - \bar{W})$ is defined as twice the deviation of such zygotes from the population mean. (This is not quite the same as the breeding value of fitness, if that is defined as twice the deviation of the *offspring's* fitness (e.g. Falconer, 1985). The parental genome would then have gone through one round of recombination, so that the breeding value depends on recombination rates.) Then:

$$\frac{W^*}{\bar{W}} = 1 + \sum_U a_{U,\varnothing}(\zeta_U + \zeta_U^* - 2C_U). \quad \text{(A 2)}$$

With weak selection, linkage disequilibria and the consequent selection on recombination are caused by deviations from multiplicative gene effects (Barton, 1995). It is therefore appropriate to work with the distribution of $\log(W^*)$, rather than of relative fitnesses, $W^*/\bar{W}$. The mean and variance of log fitness, defined for the population of newly formed zygotes, is found by taking the logarithm of eqn A 2, expanding in a Taylor's Series, and dropping terms $O(s^5)$:

$$\overline{\log(W^*/\bar{W})} = -\sum_j a_{j,\varnothing}^2 p_j q_j - 2\sum_{j<k}^* a_{j,\varnothing} a_{k,\varnothing} C_{jk} + O(s^5) \quad \text{(A 3}a\text{)}$$

$$\text{var}((\log(W^*)) = 2\sum_j a_{j,\varnothing}'^2 p_j q_j + 2\sum_{j\neq k} a_{j,\varnothing} a_{k,\varnothing} C_{jk} + 2\sum_{j<k}^* \epsilon_{jk}^2 p_j q_j p_k q_k + 2\sum_{|U|>2}^* (|U|!a_{U,\varnothing})^2 p_U q_U + O(s^5), \quad \text{(A 3}b\text{, Barton, 1995, 13}b\text{)}$$

where $a_{j,\varnothing}' = (a_{j,\varnothing} + a_{j,\varnothing}^2(p_i - q_i)/2)$, and $\epsilon_{jk} = (2a_{jk,\varnothing} - a_{j,\varnothing} a_{k,\varnothing})$ is a measure of multiplicative epistasis. Σ^* denotes a sum which counts distinct sets once, where as Σ denotes a sum in which all permutations are counted separately. The terms in eqn A 3b correspond to the variance components in eqn 8; note that to this order of approximation, linkage dis-

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

equilibria only contribute significantly to the additive variance (the second term, v_1).

We can now find the difference in mean and variance of $\log(W^*)$ between a population with some altered linkage disequilibrium, $C'_{U,V}$, and the reference values, $C_{U,V} = C_U C_V$, defined for the base population immediately after random union of gametes. We may consider the population of genomes immediately after selection ($C'_{U,V} = C_U C_V + \Delta_s C_{U,V}$), or after many generations without selection ($C_{U,V} = 0$):

$$\delta\overline{\log(W^*)} = 2\sum_{j<k}^{*} \epsilon_{jk}(C'_{jk} - C_{jk}) + 2\sum_{|U|>2}^{*} (|U|!a_{U,\varnothing})(C'_U - C_U) + O(s^5) \quad (A\,4a)$$

$$\delta\,\mathrm{var}(\log(W^*)) = 2\sum_{j\neq k} a_{j,\varnothing} a_{k,\varnothing}(C'_{jk} - C_{jk}) + O(s^5). \quad (A\,4b)$$

Here, $C_{U,\varnothing}$ has been abbreviated to C_U; because selection acts on diploid viability, $C_{U,\varnothing} = C_{\varnothing,U}$. Note that to the order of approximation used here, $\delta\,\mathrm{var}(\log(W^*)) \approx \delta\,\mathrm{var}(W^*)/\bar{W}^2$, but $\delta\overline{\log(W^*)} \approx \delta\bar{W}/\bar{W} - \delta\,\mathrm{var}(W^*)/(2\bar{W}^2)$.

In order to relate these expressions to the components of fitness variation (eqns. 8, A 3*b*), we must approximate the linkage disequilibria by assuming that selection is weak relative to recombination, so that they have reached quasi-linkage equilibrium. From Barton & Turelli (1991, eqn 25):

$$C_U = \frac{|U|!\tilde{a}_{U,\varnothing} p_U q_U}{r_U} + O(s^3). \quad (A\,5)$$

Here, r_U is the total rate of recombination among the set of loci U. First, compare a population with no linkage disequilibrium ($C'_{U,V} = 0$) with the base population. The mean decreases by:

$$\delta\overline{\log(W^*)} = -2\sum_{j<k}^{*} \frac{\epsilon_{jk}^2 p_j q_j p_k q_k}{r_{jk}} - 2\sum_{|U|>2}^{*} \frac{(|U|!a_{U,\varnothing})^2 p_U q_U}{r_U} + O(s^5) = -\sum_{k>1} V_k E\left[\frac{1}{r_U}\right]. \quad (A\,6a,\ \text{Barton, 1995, eqn 15}b)$$

The sums in eqn A 6*a* are over all distinct sets of loci, U, with two or more members. $E[1/r_U]$ is the harmonic mean rate of recombination events that disrupt sets of size $|U| = k$.

The variance changes by:

$$\delta\,\mathrm{var}(\log(W^*)) = -2\sum_{j\neq k} \frac{a_{j,\varnothing} a_{k,\varnothing} \epsilon_{jk}^2 p_j q_j p_k q_k}{r_{jk}} + O(s^5) = v_1. \quad (A\,6b)$$

By definition, this equals v_1, the contribution of pairwise linkage disequilibria to the additive genetic variance.

Published experiments have compared chromosomes extracted immediately after selection with those allowed to undergo one round of recombination. Hence, $C'_U = C_U + \Delta_s C_U$, and $C'_{U,V} = C_U C_V + \Delta_s C_{U,V}$. Substituting into eqns A 4, and noting that at quasi-linkage equilibrium, with selection on diploid viability, $C_U = \Delta_s C_U / r_U$:

$$\delta\overline{\log(W^*)} = -2\sum_{j<k}^{*} \epsilon_{jk}^2 p_j q_j p_k q_k - 2\sum_{|U|>2}^{*} (|U|!a_{U,\varnothing})^2 p_U q_U + O(s^5) = -\sum_{k>1} V_k. \quad (A\,7a)$$

The variance changes by:

$$\delta\,\mathrm{var}(\log(W^*)) = -2\sum_{j\neq k} a_{j,\varnothing} a_{k,\varnothing} \epsilon_{jk}^2 p_j q_j p_k q_k + O(s^5). = v_1 E[r_{jk}] \quad (A\,7b)$$

Equations A 7*a*, A 7*b* should be multiplied by a factor of 2 if the comparison is between chromosomes that have been extracted from males and females in *Drosophila*, as above.

The above expressions are based on the assumption that epistasis is weak relative to directional selection. If all the selection coefficients ($a_{j,\varnothing}, a_{jk,\varnothing} \ldots$ etc.) are of the same order, $\approx s$, then the change in variance in log (fitness) is small relative to the change in the mean. It is hard to find a more accurate expression for the change in variance of log(fitness) by taking into account the next order of approximation, though this can be done for the specific Gaussian forms of selection considered above. However, the effect of recombination on the mean and variance of fitness itself can be calculated exactly:

$$\frac{\delta\bar{W}^*}{\bar{W}} = \sum_{|U|>2}^{*} (|U|!a_{U,\varnothing})(C'_U - C_U) \quad (A\,8a)$$

$$\frac{\delta\,\mathrm{var}(W^*)}{\bar{W}^2} = \left(\frac{\delta\bar{W}}{\bar{W}}\right)^2 + 2\sum_U \sum_V a_{U,\varnothing} a_{V,\varnothing}(C'_{UV} + C'_{U,V} - 2C'_U C'_V - C_{UV} + C_U C_V). \quad (A\,8b)$$

Here, recombination has changed the linkage disequilibria from the reference values C_U to new values C'_U. Substitution of the quasi-linkage equilibrium approximation (eqn A 5) shows that as before, the change in mean fitness is proportional to the non-additive variance in fitness (cf. eqns A 6*a*, A 7*a*). The change in variance (eqn A 8*b*) is not obviously related to the components of fitness variation, and it is not clear that it is necessarily positive or negative. However, since terms in the sum with $U = V$ make no contribution, and since the linkage disequilibria are of order s, $\delta\,\mathrm{var}(W^*)/\bar{W}^2$ is of order s^3. This is small

https://doi.org/10.1017/S0016672300033450 Published online by Cambridge University Press

relative to the change in mean, which is of order s^2, and relative to the total genetic variance, which is also of order s^2.

References

Altenberg, L. & Feldman, M. W. (1987). Selection, generalized transmission and the evolution of modifier genes. I. The reduction principle. *Genetics* **117**, 559–572.

Baker, B. S., Carpenter, A. T. C., Esposito, M. S., Esposito, R. E. & Sandler, L. (1976). The genetic control of meiosis. *Annual Review of Genetics* **10**, 53–134.

Barton, N. H. (1995). A general theory for the evolution of recombination. *Genetical Research* **65**, 123–144.

Barton, N. H. & Turelli, M. (1991). Natural and sexual selection on many loci. *Genetics* **127**, 229–255.

Begun, D. J. & Aquadro, C. F. (1992). Levels of naturally occurring DNA polymorphism correlate with recombination rate in *Drosophila melanogaster*. *Nature* **356**, 519–520.

Berg, P. & Howe, M. M. (1989). *Mobile DNA*. Washington, D.C.: American Society for Microbiology.

Bernstein, H., Hopf, F. A. & Michod, R. E. (1988). Is meiotic recombination an adaptation for repairing DNA, producing genetic variation, or both? In *The Evolution of Sex* (ed. R. E. Michod and B. R. Levin), pp. 139–160. Sunderland, Massachusetts: Sinauer Press.

Brooks, L. D. (1988). The evolution of recombination rates. In *The Evolution of Sex* (ed. R. E. Michod and B. R. Levin), pp. 87–105. Sunderland, Massachusetts: Sinauer Press.

Bulmer, M. G. (1985). *The Mathematical Theory of Quantitative Genetics*. Oxford: Oxford University Press.

Charlesworth, B. (1976). Recombination modification in a fluctuating environment. *Genetics* **83**, 181–195.

Charlesworth, B. (1985). The population genetics of transposable elements. In *Population Genetics and Molecular Evolution* (ed. T. Ohta and K. Aoki), pp. 213–232. Berlin: Springer-Verlag.

Charlesworth, B. (1987). The heritability of fitness. In *Sexual Selection: Testing the Alternatives* (ed. J. W. Bradbury and M. B. Andersson), pp. 21–40. Chichester: John Wiley.

Charlesworth, B. (1989). The evolution of sex and recombination. *Trends in Ecology & Evolution* **4**, 264–267.

Charlesworth, B. (1990). Mutation-selection balance and the evolutionary advantage of sex and recombination. *Genetical Research* **55**, 199–221.

Charlesworth, B. (1993). Directional selection and the evolution of sex and recombination. *Genetical Research* **61**, 205–224.

Charlesworth, B. & Charlesworth, D. (1975). An experiment on recombination load in *Drosophila melanogaster*. *Genetical Research* **25**, 267–274.

Charlesworth, B. & Langley, C. H. (1989). The population genetics of *Drosophila* transposable elements. *Annual Reviews of Genetics* **23**, 251–287.

Charlesworth, B., Lapid, A. & Canada, D. (1992). The distribution of transposable elements within and between chromosomes in a population of *Drosophila melanogaster*. II. Inferences on the nature of selection against elements. *Genetical Research* **60**, 115–130.

Coyne, J. A., Meyers, W., Crittenden, A. P. & Sniegowski, P. (1993). The fertility effects of pericentric inversions in *Drosophila melanogaster*. *Genetics* **134**, 487–496.

Crow, J. F. (1970). Genetic loads and the cost of natural selection. In *Mathematical Topics in Population Genetics* (ed. K. I. Kojima), pp. 128–177. Berlin: Springer Verlag.

Davis, P. S., Shen, M. W. & Judd, B. H. (1987). Asymmetrical pairing of transposons in and proximal to the white locus of *Drosophila* accounts for four classes of regularly occurring exchange products. *Proceedings of the National Academy of Science U.S.A.* **84**, 174–178.

Dobzhansky, T., Levene, H., Spassky, B. & Spassky, N. (1959). Release of genetic variability through recombination. III. *Drosophila prosaltans*. *Genetics* **44**, 75–92.

Eanes, W. F., Wesley, C. & Charlesworth, B. (1992). Accumulation of P elements in minority inversions in natural populations of *Drosophila melanogaster*. *Genetical Research* **59**, 1–9.

Falconer, D. S. (1985). A note on Fisher's 'average effect' and 'average excess'. *Genetical Research* **46**, 337–348.

Feldman, M. W., Christiansen, F. B. & Brooks, L. D. (1980). Evolution of recombination in a constant environment. *Proceedings of the National Academy of Sciences U.S.A.* **77**, 4838–4841.

Felsenstein, J. (1974). The evolutionary advantage of recombination. *Genetics* **78**, 737–756.

Fisher, R. A. (1930). *The Genetical Theory of Natural Selection*. Oxford: Oxford University Press.

Hamilton, W. D. (1980). Sex vs non-sex vs parasite. *Oikos* **35**, 282–290.

Hamilton, W. D. (1993). Haploid dynamic polymorphism in a host with matching parasites: effects of mutation/subdivision, linkage and patterns of selection. *Journal of Heredity* **84**, 328–338.

Houle, D. (1992). Comparing evolvability and variability of quantitative traits. *Genetics* **130**, 195–204.

Keightley, P. D. (1994). The distribution of mutation effects on viability in *Drosophila*. *Genetics* **138**, 1315–1322.

Kimura, M. & Maruyama, T. (1966). Mutational load with epistatic gene interactions in fitness. *Genetics* **54**, 1337–1351.

Kondrashov, A. S. (1984). Deleterious mutations as an evolutionary factor. I. The advantage of recombination. *Genetical Research* **44**, 199–218.

Kondrashov, A. S. (1995). Dynamics of unconditionally deleterious mutations: Gaussian approximation and soft selection. *Genetical Research* **65**, 113–122.

Langley, C. H., Montgomery, E., Hudson, R. Kaplan, N. & Charlesworth, B. (1988). On the role of unequal exchange in the containment of transposable element copy number. *Genetical Research* **52**, 223–235.

Levene, H. (1959). Release of genetic variability through recombination. IV. Statistical theory. *Genetics* **44**, 93–104.

Maynard Smith, J. (1978). *The Evolution of Sex*. Cambridge: Cambridge University Press.

Maynard Smith, J. (1980). Selection for recombination in a polygenic model. *Genetical Research* **33**, 121–128.

Maynard Smith, J. (1988). Selection for recombination in a polygenic model: the mechanism. *Genetical Research* **51**, 59–63.

Montgomery, E. A., Huang, S. M., Langley, C. H. & Judd, B. H. (1991). Chromosome rearrangement by ectopic recombination in *Drosophila melanogaster*: genome structure and evolution. *Genetics* **129**, 1085–1098.

Mukai, T. (1969). The genetic structure of natural populations of *D. melanogaster*. VIII. Synergistic interactions of spontaneous mutant polygenes controlling viability. *Genetics* **61**, 149–161.

Mukai, T. & Yamaguchi, O. (1974). The genetic structure of natural populations of *D. melanogaster*. XI. Genetic variability in a local population. *Genetics* **76**, 339–366.

Nee, S. (1989). Antagonistic coevolution and the evolution of genotypic randomisation. *Journal of Theoretical Biology* **140**, 499–518.

Nuzhdin, S. V. & Mackay, T. F. C. (1995). The genomic rate of transposable element movement in *Drosophila melanogaster*. *Molecular Biology and Evolution* **12**, 180–181.

Radman, M. & Wagner, M. (1993). Mismatch recognition in chromosomal interactions and speciation. *Chromosoma* **102**, 369–373.

Shnol, E. E. & Kondrashov, A. S. (1993). The effect of selection on the phenotypic variance. *Genetics* **134**, 995–996.

Sniegowski, P. D. & Charlesworth, B. (1994). Transposable element numbers in cosmopolitan inversions from a natural population of *Drosophila melanogaster*. *Genetics* **137**, 815–827.

Spassky, B., Spassky, N., Levene, H. & Dobzhansky, T. (1958). Release of genetic variability through recombination. I. *Drosophila pseudoobscura*. *Genetics* **43**, 844–865.

Spiess, E. B. (1958*a*). The effect of genetic recombination on viability in *Drosophila*. *Cold Spring Harbor Symposium on Quantitative Biology* **23**, 239–250.

Spiess, E. B. (1958*b*). Release of genetic variability through recombination. II. *Drosophila persimilis*. *Genetics* **44**, 43–58.

Tucíc, N., Ayala, F. J. & Marinkovic, D. (1981). Correlation between recombination frequency and fitness in *Drosophila melanogaster*. *Genetica* **56**, 61–69.

Turelli, M. (1984). Lerch's zeta meets the abdominal bristle. *Theoretical Population Biology* **25**, 138–193.

Turner, J. R. G. (1967). Why does the genotype not congeal? *Evolution* **21**, 645–656.

Wasserman, M. (1972). Factors influencing fitness in chromosomal strains in *Drosophila subobscura*. *Genetics* **72**, 691–708.

Woodruff, R. C., Slatko, B. E. & Thompson, J. R. (1983). Factors affecting mutation rate in natural populations. In *The Genetics and Biology of Drosophila*, vol. 3c (ed. M. Ashburner, H. L. Carson and J. N. Thompson), pp. 37–124. London: Academic Press.

Zhivotovsky, L. A., Feldman, M. W. & Christiansen, F. B. (1994). Evolution of recombination among multiple selected loci: a generalized reduction principle. *Proceedings of the National Academy of Sciences U.S.A.* **91**, 1079–1083.