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Maintenance of genetic homogeneity in systems with multiple genomes

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SUMMARY

Genes or sequences of DNA present in multiple copies per cell include entire genomes of mitochondria and chloroplasts, nuclear ribosomal RNA genes, and highly repetitive sequences in heterochromatin. All copies are nearly identical, in spite of mutational pressure and weak selection. A zygote containing mitochondrial or chloroplast genophores of two different genotypes quickly produces progeny pure for one genotype or another (vegetative segregation). Evidence from yeast and *Chlamydomonas* suggests that organelle genophores undergo repeated rounds of random mating and recombination. When two molecules carrying different alleles at a locus recombine, gene conversion can result in the cell becoming pure for one allele. Stochastic matching and conversion (SMAC) has been studied by computer simulations which suggest that it will tend to eliminate new mutations in yeast mitochondrial DNA and speed up vegetative segregation. We have verified previous suggestions that gene conversion, occurring during unequal mitotic sister-strand crossing-over, provides an efficient mechanism for maintaining the homogeneity of repeated sequences in eukaryotic chromosomes.

1. INTRODUCTION

The laws of classical genetics are designed principally to describe the behaviour of genes which are present in only one to four copies per cell, one on each chromosome, so that each gene has a limited number of pairing partners for recombination. In contrast, the DNA molecules of mitochondria and chloroplasts, which we shall call genophores hereafter to avoid confusion with true chromosomes, are present in many copies in each cell. Each genophore, and consequently each gene, has many potential partners for recombination. In addition, it has recently been clearly established that many nucleotide sequences on chromosomes, such as those in heterochromatin or the genes coding for ribosomal RNA, are present in multiple copies in tandem arrays. To the extent that unequal crossing-over occurs, each such sequence also has a number of different recombination partners.

There is now good genetic evidence that mitochondrial genophores in at least one organism, *Saccharomyces cerevisiae*, undergo repeated rounds of random mating (pairing and recombination) in a manner analogous to the phage-infected bacterial cell (Visconti & Delbrück, 1953). The evidence has been provided mainly by Dujon and co-workers (1974, 1975) and verified by Birky & Perlman (unpublished) and

others (reviewed by Birky, 1975). Dujon *et al.* estimated from different sets of data that each gene on each genophore undergoes, on the average, 3·7 (1974) or 4·6 (1975) rounds of mating (heteroduplex formation and repair) in a zygote and its progeny before vegetative segregation is complete; this probably amounts to at least one or two rounds of mating per cell generation. Multiple rounds of mating are also suggested by the results of density labelling experiments with diploid yeast (Williamson & Fennell, 1974).

Recent studies on the recombination of chloroplast genes in *Chlamydomonas* are also interpreted in terms of a random mating pool of organelle genophores (Gillham, Boynton & Lee, 1974 and personal communication). Recombination of mitochondrial genes has been searched for and found in *Aspergillus* (Rowlands & Turner, 1974) and in *Paramecium* (Adouette, 1974); the genetic analysis in other systems is not sophisticated enough to detect it, but observations of repeated fusion and fission of organelles in many organisms suggest that random mating pools will prove to be a widespread phenomenon. Other types of genetic system which potentially involve random mating of multiple genophores include intracellular symbionts and highly polyploid nuclei such as the macronuclei of ciliates, non-pathogenic viruses, and to a first approximation, repetitive DNA in chromosomes (as we will argue in section 5 below).

It is now generally accepted that recombination in both prokaryotes and eukaryotes involves the formation of heteroduplex DNA molecules, and that the repair of mismatched base pairs in those heteroduplex regions often leads to gene conversion, which may or may not be accompanied by classical crossing-over (reviewed by Radding, 1973; Meselson & Radding, 1975). As we illustrate in Fig. 1, there are four possible outcomes of a recombination event at a particular locus: two of the events result in a detectable conversion, with the mating molecules becoming *genetically identical* at that locus. Now consider a population of genophores or chromosomes, initially heterogeneous at a particular locus, undergoing random mating with gene conversion. It is apparent that the gene frequency in the population will drift randomly; repeated rounds of mating will sooner or later result in the frequency of any particular allele reaching 1 or 0, i.e. the population will become genetically homogenous at the locus in question and, in time, at every locus. This random drift toward the fixation of one allele or the other does not require that mating be *completely* random; to avoid implying that it does, we refer to the process as *stochastic matching and conversion*, and use the acronym SMAC.

SMAC is potentially a force for the establishment and maintenance of genetic homogeneity in systems having multiple copies of genophores or of single genes. Its effectiveness depends upon the rate of approach to homogeneity, relative to opposing forces such as mutation. In this paper we will describe the results of computer simulations of SMAC. In view of the results, we argue that it may play an important role in eliminating new mutations in organelle genes, in acclerating the rate of vegetative segregation of organelle genes, and in the rectification of repetitive DNA sequences in chromosomes. Some of this work has already been briefly presented (Birky & Skavaril, 1975).

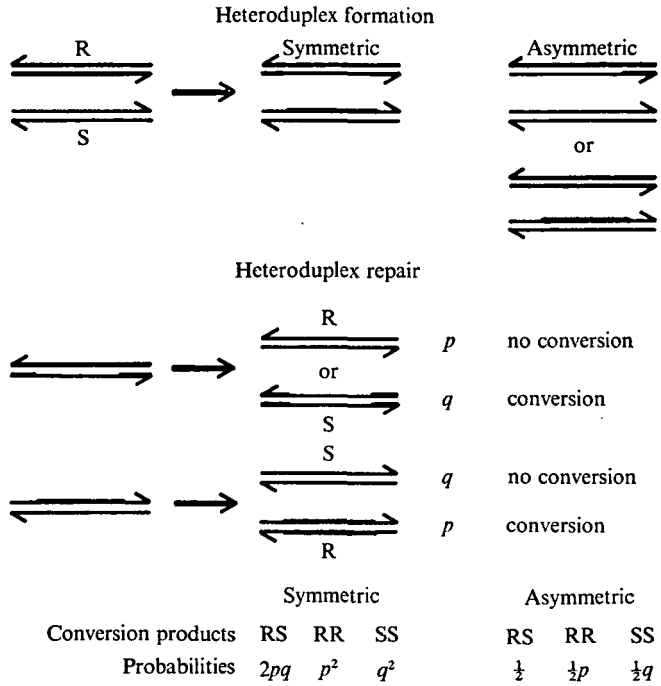

Fig. 1. Gene conversion. Alleles are designated R and S. Heteroduplex formation may be symmetric or asymmetric. In either case, heteroduplex repair can result in conversion or no conversion. We have assumed symmetric heteroduplex formation, in which case the probability that two molecules become identical is p^2+q^2, where p is the probability that the R strand is copied during repair, and q is the probability that the S strand is copied.

2. METHODS

The stochastic computer simulation programs were written in Conversational Programming System (CPS) language and executed on an IBM 370/165. Two models were simulated: (i) random mating of unlinked genes on organelle genophores; and (ii) unequal sister-strand recombination among linked multiple gene copies.

For the random mating model, the parameters of the simulation were the initial number of genophores carrying the R and S alleles; the probabilities of no conversion, conversion of RS to RR, conversion of RS to SS, and conversion of RS to SR; the number of rounds of mating to be simulated; and the number of replicate simulations (runs) to be performed. Each run began with the establishment of an initial gene array, according to the simulation parameters. Within each round of mating, all the genes were paired at random; and gene conversion was applied stochastically to those pairs of genes whose configuration permitted heteroduplex formation (either RS or SR), according to the conversion probability parameters. This process was repeated for the number of cycles of mating specified, or until homogeneity of the gene array had been achieved, whereupon the program wrote the number of cycles of mating that had been simulated and the percentage of the R type genes present in the final gene array. In effect, each such run represents the fate

of one cell. We then formed cumulative frequency distributions from the data from many runs.

We have assumed that heteroduplex formation during recombination is symmetric (see Fig. 1); and, unless otherwise specified, that there is no preferential direction of conversion, i.e. that an R × S mating will have four, equally probable consequences, with associated probabilities each equal to 0·25: (i) RS (no conversion); (ii) SR (both alleles converted); (iii) RR (S allele converted); and (iv) SS (R allele converted). Asymmetric heteroduplex formation would have given the same probabilities when the two conversion rates are each 0·5, but it gives a lower total probability of conversion when the two conversion rates are unequal.

For the simulation of linked multiple copies of genes (cf. Fig. 5 and text, section 5), the input parameters were the number of tandemly linked multiple copies of the gene, the maximum latitude allowed at chromatid alignment, the number of cycles of mating rounds to be simulated, and the number of runs to be performed. The initial position of a mutant allele could be set either by means of an input parameter, or established at random by the program. During each cycle of mating in this model, a random alignment of the sister chromatids was simulated, subject to the constraint imposed by the maximum latitude allowed for misalignment. Next, a site for possible gene conversion was selected at random. If the site represented a heteroduplex configuration, then one of the four possible gene conversion consequences was selected at random and applied to the site selected. The process of site selection and possible gene conversion for a given chromatid alignment could be set to be simulated more than once per cycle of mating, if desired. The cycle of mating was concluded with one chromatid being selected at random for replication. The next cycle of mating was applied to the replicated pair of chromatids, the non-replicated chromatid being discarded. This process was continued for the number of mating cycles specified, or until homogeneity of the genome had been achieved, whichever happened first. At the end of each simulation replicate, the program wrote the number of rounds of mating that had been applied and the numbers of each of the two types of allele present in the final genome.

3. THE FATE OF NEW MUTATIONS IN YEAST MITOCHONDRIAL DNA

A haploid cell of *Saccharomyces cerevisiae* contains approximately 50 molecules of mitochondrial DNA (mtDNA); all molecules carry the same set of gene loci, so that the cell is effectively 50-ploid. It is extremely unlikely that a point mutation would occur in the same gene in more than one of these molecules at a time. (Our discussion will not deal with *petite* mutations, which involve extensive deletions and duplications of the mitochondrial genome and which may spread throughout the population by *directed* gene conversion: Perlman & Birky, 1974.) Moreover, for most of the commonly studied types of mitochondrial mutation, such as those which confer antibiotic resistance on mitochondrial ribosomes, it is unlikely that a single mutant gene will be phenotypically expressed.

This situation is simulated in the computer by starting with a population of 49

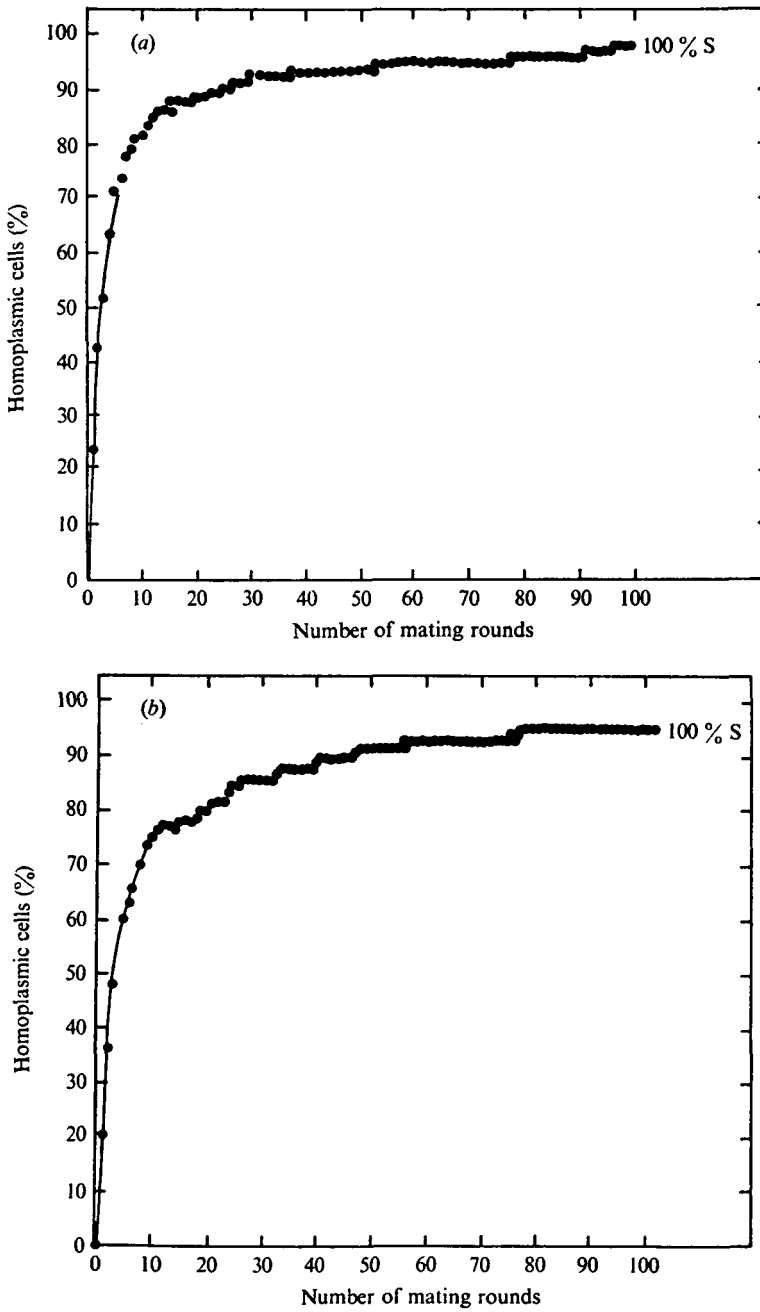

Fig. 2. The results of 100 simulations of SMAC, corresponding to 100 cells, with inputs (*a*) 1R:49S, and (*b*) 1R:99S. Data are shown as cumulative frequencies of genetically homogeneous cells for the first 100 rounds of mating. In (*a*), all cells were genetically homogeneous (homoplasmic) after the 312th round: one cell became 100% R after round 312. In (*b*), all cells were homoplasmic after the 595th round; one cell became 100% R in round 154.

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wild type, antibiotic-sensitive (S) genes and one mutant, resistant (R) gene. These genes are paired randomly. Pairs of like genes (RR and SS pairs) remain unchanged, while an RS pair has a probability of 0·25 of becoming RR and 0·25 of becoming SS. At the end of this first round of mating, the genes are paired randomly again and the process repeated, until such time as the entire population becomes homogeneous (homoplasmic, for organelle genes) for the S or the R allele. This simulation was repeated 100 times, to simulate events in 100 different cells. The cumulative frequency distribution is shown in Fig. 2(*a*). The majority of the cells become homoplasmic for the S allele; 24% in one round of mating, 43% in two rounds. This would correspond to approximately one cell generation. Theoretically, 2% of the cells should by chance become homogeneous for the mutant R allele, very slowly; in these 100 simulations one cell fixed the R allele after 312 mating rounds.

These simulations clearly do not provide a realistic picture of events in the yeast cell after the first few rounds of mating because they do not take into account the influence of mtDNA replication, of segregation of mtDNA molecules during budding, or of the packaging of mtDNA molecules into mitochondria. We have begun to develop a program which will include these features. Replication of organelle genophores will slow the approach to genetic homogeneity by increasing the total number of interacting organelles, but this effect will be small, as can be seen by comparing the results of computer simulations of SMAC in a diploid cell with 1 R and 99 S molecules (Fig. 2*b*). In that case, 21% of the cells lost the mutant allele in one round, 37% in two rounds. Packaging of several mtDNA molecules into each mitochondrion may speed up the approach to homogeneity if there is a tendency for molecules to mate with others in the same mitochondrion, as this will reduce the effective population size, but this effect will also be small (cf. Fig. 3*b* below). Since segregation by itself could result in the production of homozygous cells, it is likely to speed up the process considerably (see section 4 below). Perhaps most importantly, intracellular selection for the wild-type allele (Birky, 1973) may intervene and speed up the loss of mutant alleles.

Our conclusion, then, is that a large proportion of new mutant alleles will be lost by gene conversion during the first generation after their occurrence, unless they are rescued by intracellular selection. Experimental tests of this prediction are under way in yeast. The same prediction applies to other systems containing multiple genomes, especially in *Chlamydomonas* where there is evidence for random mating. We see, moreover, an obvious consequence for the process of evolution in such systems: a reduction in the effective mutation rate, i.e. in the rate of production of cells showing the mutant phenotype.

4. VEGETATIVE SEGREGATION OF YEAST MITOCHONDRIAL GENES

The experimental analysis of mitochondrial genetics in yeast (reviewed by Birky, 1975) begins with the fusion of haploid cells differing in mitochondrial genotype (e.g. R × S) to form a zygote which initially is heteroplasmic (genetically heterogeneous). Beginning even in the first generation, vegetative reproduction

produces a high proportion of diploid progeny which are homoplasmic (pure) for one genotype or the other, and all cells in the poulation are homoplasmic within 5–10 generations. A classical explanation for this process of vegetative segregation is that organelle genes are distributed randomly between mother and bud, resulting in the gradual sorting-out of genophores of different genotypes. Complete sorting-out takes about $10N$ generations, where N is the number of segregating units immediately after cell division (Michaelis, 1955).

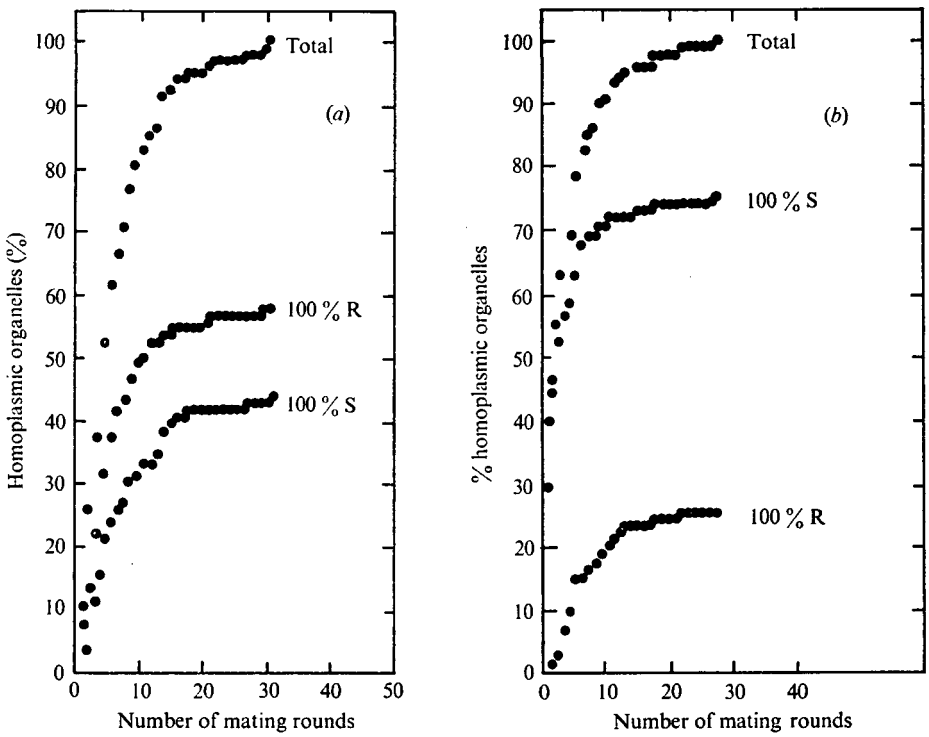

Fig. 3. Results of 100 simulations of SMAC with inputs (*a*) 2R:2S, and (*b*) 1R:3S.

In yeast, newly formed zygotes will contain at least 100 molecules of mtDNA. If the mitochondrial genophore were the unit of segregation, complete sorting-out would take at least 1000 generations. Dujon *et al.* (1974) determined the frequency with which the first buds formed by zygotes were homozygous, and from this datum calculated the number of segregating units to be about three, assuming random segregation. They postulated that only three mitochondrial genophores enter the bud, the remainder staying in the mother zygote. But this is unlikely, for reasons discussed by Birky (1975) and Callen (1974); recent data indicate that about 20 mitochondrial genophores enter the bud (Birky & Strausberg, unpublished).

The number of mitochondria in a yeast cell is generally much smaller than the number of mtDNA molecules, and under some circumstances may be on the order of magnitude of the number of segregating units (e.g. Grimes, Mahler & Perlman,

1974). But if the segregating unit is the mitochondrion, then all of the mitochondrial genophores in each organelle must be genetically identical. This is not expected, since mitochondria from the haploid parents must presumably fuse to permit recombination. Birky (1975) postulated that the mitochondrial genophores in such mitochondria undergo repeated rounds of SMAC, tending to maintain genetic homogeneity inside each organelle.

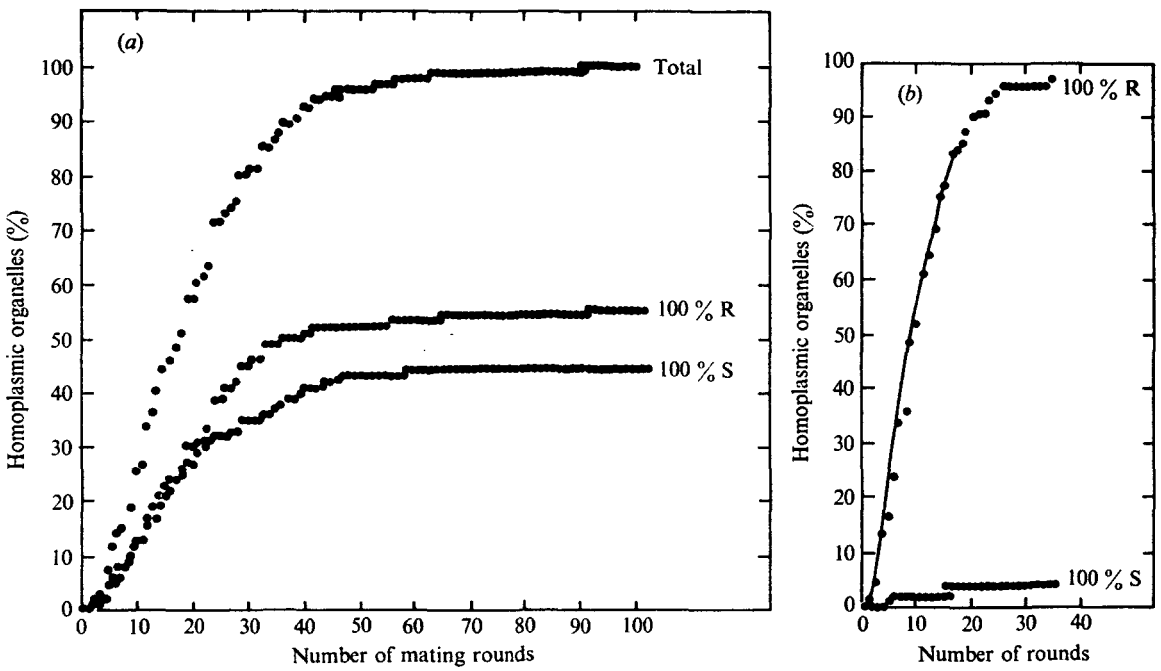

Fig. 4. Results of 100 simulations of SMAC with input of 5R:5S. The probabilities of heteroduplex repair, p and q, were 0·5 in (*a*), and 0·6 and 0·4 in (*b*). Data are shown for the first 100 mating rounds only in (*a*); one cell became 100 % S after 135 rounds.

The results of computer simulations are compatible with this hypothesis. Fig. 3(*a*) shows that 10 % of mitochondria with four genophores, two carrying R and two carrying S alleles, will become fixed for one allele or the other in one round of SMAC; 24 % are fixed in two rounds. Most mitochondria will, by chance, have unequal numbers of R and S genophores, which will speed up the approach to homogeneity appreciably. Fig. 3(*b*) shows that 26 % of mitochondria with one R and three S genes are fixed in one round, and 46 % are genetically homogeneous in two rounds of mating. Larger mitochondria drift substantially more slowly; with five R and five S genes, five rounds are required to achieve homogeneity in 7 % of the cases (Fig. 4*a*).

There is evidence from several genetic systems that the rate of gene conversion from the wild type to the mutant allele may differ from the rate from mutant to wild type (in the terminology of Fig. 1, $p \neq q$). Preferential gene conversion of this sort is illustrated for a population of five R and five S genes in Fig. 4(*b*) ($p = 0{\cdot}6$,

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$q = 0{\cdot}4$). Preferential gene conversion has two striking effects: (i) it drastically increases the rate of fixation of alleles (cf. Figs. 4*a*, *b*); and (ii) it biases the ratio of cells becoming homozygous for R and S strongly in favour of one allele (96% R in the case illustrated). Theoretically, after the first round the average frequency of the R allele should change from the initial value of 0·5 to $p^2 + pq = 0{\cdot}6$; the proportion of R alleles will increase in each subsequent generation. This biasing effect would be smaller in the case of asymmetric heteroduplexes.

We conclude that SMAC may be an effective force in maintaining the genetic homogeneity of mtDNA molecules inside individual zygote mitochondria. This in turn would tend to make the organelle, rather than the molecule, the unit of vegetative segregation. SMAC taking place in subpopulations of mtDNA molecules might also explain vegetative segregation of mitochondrial genes in *Aspergillus* (Rowlands & Turner, 1974). Cells in mycelia of this organism are not completely separated by walls, so that sorting-out cannot explain vegetative segregation.

5. THE RECTIFICATION OF REPETITIVE DNA SEQUENCES IN CHROMOSOMES

It is now well established that a substantial fraction of the DNA in eukaryotic chromosomes consists of families of similar or identical (repeated) sequences (reviewed by Tartof, 1975). In at least some instances, the sequences are arranged as tandem repeats. A thoroughly studied example is the set of sequences coding for ribosomal RNA (rRNA) in animals, present as several hundred copies, all identical or nearly so and tandemly arranged in the nucleolus organizer; each copy contains about $1{\cdot}3 \times 10^4$ nucleotide pairs (Brown & Sugimoto, 1974). At the other extreme is the highly repetitive DNA found in heterochromatin, consisting of thousands of copies of very short sequences (e.g. seven nucleotide pairs in *Drosophila virilis*, according to Gall, Cohen & Atherton, 1974). The homogeneity of these sequences seems paradoxical. Mutations in one or a few copies should be selectively neutral in the presence of hundreds or thousands of wild-type copies. Such mutations should, accordingly, accumulate in time and homogeneity should be lost. That mutation *can* occur in these sequences is shown by differences in base sequence in the rDNA of *Xenopus laevis* and *X. mulleri* (Brown & Sugimoto, 1974). There must, then, be some means for the rectification of sequences which have diverged.

A number of rectification models have been proposed. Callan (1967) suggested that one of the copies is a 'master' copy against which all the other ('slaves') are matched and corrected in each generation. Thomas (1970, 1974) proposed two different models whereby *any* copy could serve as a master. Buongiorno-Nardelli, Amaldi & Lavasanchez (1972) suggested that all copies are set free from the chromosome and amplified by rolling-circle replication; the tandemly linked copies of only one sequence are then inserted back into the chromosome. Tartof (1974) and Smith (1974) suggested that unequal crossing-over between sister chromatids during mitosis in the germ line could result in the random re-shuffling of sequences; eventually, chromatids would be produced which contained a majority of mutant sequences, which would then be exposed to selection. Inspired by this suggestion and

by our consideration of SMAC in organelle genes, we designed another model which proved to be identical to one first proposed by Edelman & Gally (1970; see also Tartof, 1975) and called by them 'democratic gene conversion'. The term 'democatic' was chosen to contrast this model, in which *any* sequence can become fixed, with the master-slave hypothesis of Callan.

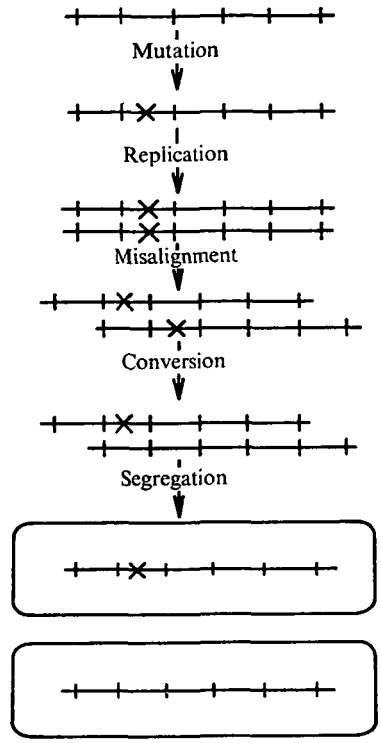

Fig. 5. The hypothesis of democratic gene conversion: SMAC during unequal mitotic sister-strand recombination. See text for explanation.

In this model, we suppose that a chromosome containing a set of tandemly repeated, identical sequences (hereafter, 'genes') has suffered a mutation in one gene. As shown in Fig. 5, this chromosome now replicates in the next S phase. Subsequently, the sister strands pair and undergo gene conversion inside the region of repeated sequences. Reciprocal sister-strand exchanges were first detected by Taylor, Woods & Hughes (1957) in autoradiographic studies; although many are induced by radiation from the tritium used in such studies, exchanges are also detectable by other techniques (Ikushima & Wolff, 1974) and may occur spontaneously. They presumably involve gene conversion as well as segmental exchange. We further suppose, as did Smith and Tartof, that sister-strand exchange in a region of tandem repeats may be unequal, i.e. that the pairing strands may be out of register by one or more repeats, as sometimes happens during meiosis. This unequal exchange introduces an element of random mating into the system. The mutation can spread to other repeats, but in general it will be eliminated since it is out-

numbered by wild-type sequences. The rate at which the mutant allele is lost or fixed would be expected to depend upon (i) the position of the initial repeat; (ii) the latitude, i.e. the maximum extent to which sister strands may be out of register; and (iii) the number of copies of the gene.

We have carried out computer simulations of this process, as described in Methods. We first studied the effect of the starting position of the mutation. A

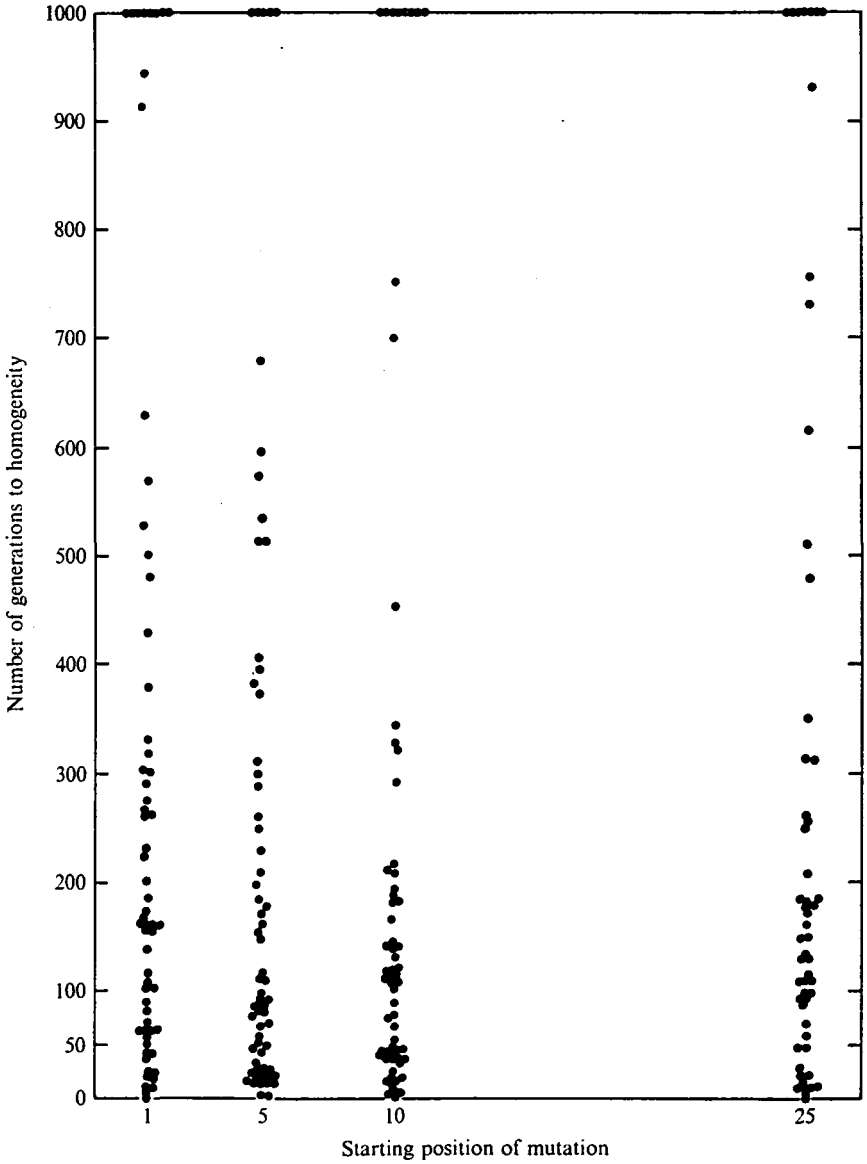

Fig. 6. Simulations of democratic gene conversion: effect of starting position of initial mutation on number of generations, with recombination, required to achieve homogeneity. For each starting position, 65 simulations were done, corresponding to 65 different chromosomes.

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series of 50 identical repeats was used, with a maximum latitude of 5 repeats (in each round of recombination, the actual degree of mismatch is chosen randomly to be between 0 and 5 genes). Recombination was permitted to occur only in the region of overlap, and involved only one gene, selected at random. Each recombination event could result in conversion or not, as described previously. After each round of replication and recombination, one daughter chromosome was selected randomly and served as the starting point for the next round. The starting position of the mutation was set as the first (end), fifth, tenth, or twenty-fifth (middle) gene in the set of 50 genes. The results are shown in Fig. 6 and summarized in Table 1. Chromosomes

Table 1. *Simulations of democratic gene conversion: summary of results*

Number of repeats	Latitude	Starting position	Generations to homogeneity: Mean	Generations to homogeneity: Standard error	Number of simulations
50	5	1	303	41	65* (8)
50	5	5	230	35	65* (5)
50	5	10	223	38	65* (8)
50	5	25	282	43	65* (9)
100	1	Random	3799	1210	20† (3)
100	5	Random	2832	1077	20† (1)
100	10	Random	907	494	30† (1)
100	15	Random	647	230	20
300	30	Random	1125	380	20
500	50	Random	472	138	20

* Simulations carried to 1000 generations only; several not finished (homogeneous) were counted as 1000 generations for calculating mean and standard error. Number not finished given in parentheses.

† Simulations carried to 15000 generations only; a few not finished were counted as 15000 generations for calculating mean and standard error. Number not finished given in parentheses.

starting with a mutation in gene position 1 approach homogeneity more slowly than others, presumably because in many rounds they will not be in the region of overlap and hence cannot recombine. There is no detectable effect of starting position between genes 5 and 25, presumably because they are always in the region of overlap. In all subsequent simulations, the starting position of the mutation was chosen randomly in each simulation, and the site of recombination was permitted to occur anywhere (though recombination outside the region of overlap would not be detectable).

The effect of latitude is shown in Fig. 7 and Table 1. The greater the permitted latitude, the faster the approach to genetic homogeneity, especially between 1% and 10% latitude. The effect of increasing the number of repeats is shown in Fig. 8 and Table 1. That effect appears to be small, and shows no consistent trend with increasing repeat number from 100 to 500. (The difference between 50 and 100 may be due to permitting recombination to occur outside the region of overlap in the latter, which would slow down the approach to homogeneity.) It might be

expected that increasing the number of repeats would have little effect, since most mutations are eliminated rather than spread, and the number of effective pairing partners for a mutant gene (the effective population size) is determined by the latitude rather than by the repeat number. In these simulations, there was one recombination event for every 100 copies, so the probability of recombination per copy remained constant.

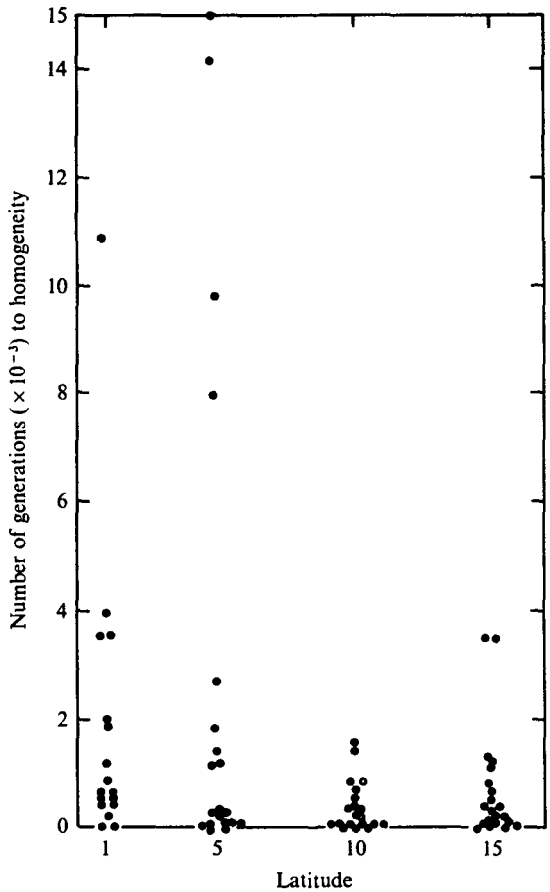

Fig. 7. Simulations of democratic gene conversion: effect of latitude. Twenty simulations were done for each latitude.

For a set of 100 genes and 10% latitude, an average of about 900 generations *with recombination* is required to rectify a chromosome. This value was calculated including one chromosome which was still not rectified when the program was stopped after 15000 generations; if this chromosome is omitted, an average of 422 generations with recombination are required. We can now calculate the actual number of cell generations required if we know the probability of recombination per generation. Ikushima & Wolff (1974) found 0·1 sister strand exchanges per chromosome per generation in cultured chinese hamster ovary cells, or 2·4 exchanges per haploid genome. Sparrow, Price & Underbrink (1972) give the genome size of the

related golden hamster as $7{\cdot}6 \times 10^7$ nucleotides per haploid genome. Accordingly, we estimate $3{\cdot}16 \times 10^{-8}$ recombination events per nucleotide pair per generation. If the genes under consideration are those coding for rRNA, we can use the value of $1{\cdot}3 \times 10^4$ nucleotide pairs per gene (from *Xenopus*) to estimate $4{\cdot}1 \times 10^{-4}$ recombination events per gene per generation. This means that 422 or 900 generations with

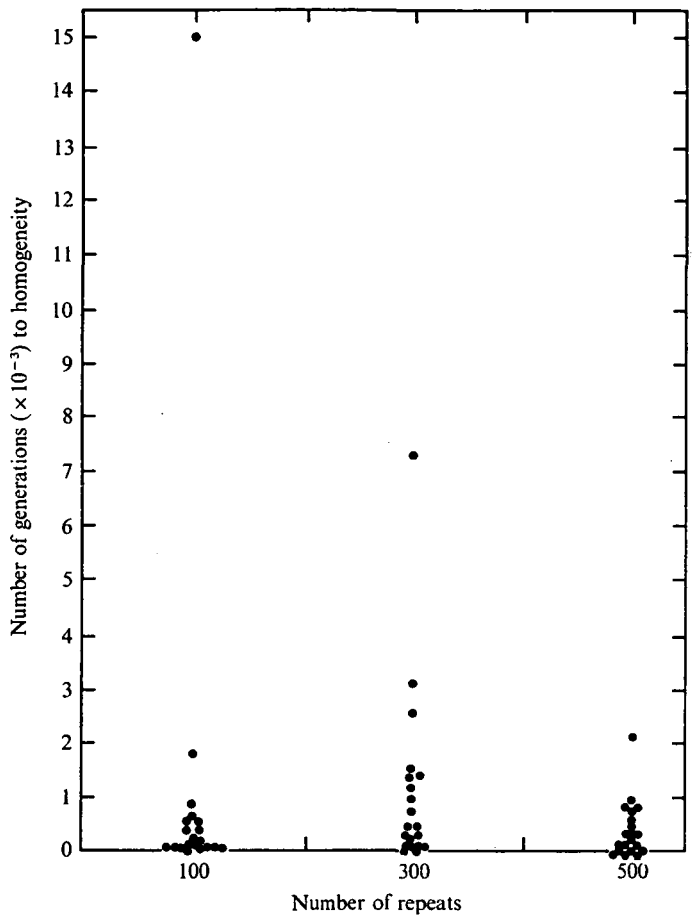

Fig. 8. Simulations of democratic gene conversion: effect of number of repeats. Twenty simulations for each number of repeats.

recombination are equivalent to a total of $1{\cdot}03 \times 10^6$ or $2{\cdot}19 \times 10^6$ total generations. How many new mutations will occur in this time? Assuming the mutation rate in *Drosophila* to be 7×10^{-11} mutations per nucleotide pair per cell generation (Drake, 1970), we can calculate that 0·94–1·8 new mutations will occur in the time it takes to eliminate one mutation. Democratic gene conversion is thus capable of eliminating new mutations in the germ line approximately as fast as they occur. Rectification might actually occur somewhat faster, since selection could intervene when the number of mutant genes becomes large enough by chance to affect the phenotype of the cell.

Democratic gene conversion differs from the model of Smith and Tartof in that the former utilizes gene conversion while the latter utilizes reciprocal exchanges of chromosome segments (crossing-over). Democratic gene conversion, unlike the Smith and Tartof model, can eliminate mutant genes from the population without any selection whatsoever, and is markedly less sensitive to repeat number, i.e. it retains its efficiency at high repeat numbers such as are encountered in heterochromatin. But democratic gene conversion will only rarely produce a chromosome homogeneous for mutant alleles, and will do it very slowly. A truly realistic model would incorporate *both* gene conversion and crossing-over, since both probably occur during recombination events; hopefully such a model would show the best features of both.

6. OTHER POSSIBLE APPLICATIONS

Two other possible applications of SMAC are under active investigation. The first of these is to the phenomenon of uniparental inheritance of organelle genes (reviewed by Birky, 1975). If a zygote receives very many more chloroplast or mitochondrial genophores from one parent (e.g. the maternal) than from the other, SMAC will rather quickly eliminate the minority alleles entirely from some zygotes. Experimental tests eliminate this as a *complete* explanation of uniparental inheritance in yeast (Birky & Demko, unpublished) and in *Chlamydomonas* (Karen Van Winkle-Swift, personal communication).

A second application might be to the phenomenon of phenotypic assortment in *Tetrahymena* (Orias & Flacks, 1975). We have been investigating two possible models: (i) random mating and conversion among the entire population of chromosomes in the macronucleus, coupled with random segregation of individual chromosomes; and (ii) recombination between chromosomes inside diploid subunits, coupled with random segregation of subunits. In both cases we have attempted to produce the observed unequal outputs with preferential gene conversion. We have had no success with the first model, but with the second model have managed to mimic the actual kinetics of phenotypic assortment closely but not exactly.

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REFERENCES

Adoutte, A. (1974). Mitochondrial mutations in Paramecium: phenotypical characterization and recombination. *The Biogenesis of Mitochondria* (ed. A. M. Kroon and C. Saccone), pp. 263–271. New York: Academic Press, Inc.

Birky, C. W., Jr. (1973). On the origin of mitochondrial mutants: evidence for intracellular selection of mitochondria in the origin of antibiotic-resistant cells in yeast. *Genetics* **74**, 421–432.

Birky, C. W., Jr. (1975). Mitochondrial genetics in fungi and ciliates. *Genetics and Biogenesis of Mitochondria and Chloroplasts* (ed. C. W. Birky, Jr., P. S. Perlman, and T. J. Byers), pp. 182–224. Columbus: Ohio State University Press.

Birky, C. W., Jr. & Skavaril, R. V. (1975). A possible role for gene conversion in achieving and maintaining genetic homogeneity among organelle genophores and repeated DNA sequences in general. *Genetics* **80**, s 14.

Brown, D. B. & Sugimoto, K. (1974). The structure and evolution of ribosomal and 5S DNAs in *Xenopus laevis* and *Xenopus mulleri*. *Cold Spring Harbor Symposia on Quantitative Biology* **38**, 501–505.

Buongiorno-Nardelli, M., Amaldi, F. & Lavasanchez, P. (1972). Amplification as a rectification mechanism for the redundant rRNA genes. *Nature New Biology* **238**, 134–137.

Callan, H. G. (1967). The organization of genetic units in chromosomes. *Journal of Cell Science* **2**, 1–7.

Callen, D. F. (1974). Segregation of mitochondrially inherited antibiotic resistance genes in zygote cell lineages of *Saccharomyces cerevisiae*. *Molecular and General Genetics* **104**, 65–76.

Catcheside, D. G. (1974). Fungal genetics. *Annual Review of Genetics* **8**, 279–300.

Drake, J. W. (1970). *The Molecular Basis of Mutation*. San Francisco: Holden-Day.

Dujon, B., Slonimski, P. P. & Weill, L. (1974). Mitochondrial genetics. IX. A model for recombination and segregation of mitochondrial genomes in *Saccharomyces cerevisiae*. *Genetics* **78**, 415–437.

Dujon, B., Kruszewska, A., Slonimski, P. P., Bolotin-Fukuhara, M., Coen, D., Deutsch, J., Netter, P. & Weill, L. (1975). Mitochondrial genetics. X. Effects of UV irradiation on transmission and recombination of mitochondrial genes in *Saccharomyces cerevisiae*. *Molecular and General Genetics* **137**, 29–72.

Edelman, G. M. & Gally, J. A. (1970). Arrangement and evolution of eukaryotic genes. *The Neurosciences: Second Study Program* (ed. F. O. Schmitt), pp. 962–972. New York: Rockefeller University Press.

Gall, J. G., Cohen, E. H. & Atherton, D. D. (1974). The satellite DNAs of *Drosophila virilis*. *Cold Spring Harbor Symposia on Quantitative Biology* **38**, 417–427.

Gillham, N. W., Boynton, J. E. & Lee, R. W. (1974). Segregation and recombination of non-Mendelian genes in *Chlamydomonas*. *Genetics* **78**, 439–457.

Grimes, G. W., Mahler, H. R. & Perlman, P. S. (1974). Nuclear gene dosage effects on mitochondrial mass and DNA. *Journal of Cell Biology* **61**, 565–574.

Ikushima, T. & Wolff, S. (1974). Sister chromatid exchanges induced by light flashes to 5-bromodeoxyuridine- and 5-iododeoxyuridine substituted chinese hamster chromosomes. *Experimental Cell Research* **87**, 15–19.

Meselson, M. S. & Radding, C. M. (1975). A general model for genetic recombination. *Proceedings of the National Academy of Sciences, U.S.A.* **72**, 358–361.

Michaelis, P. (1955). Über Gesetzmässigkeiten der Plasmon-Umkonbination und über eine Methode zur Trennung einer Plastiden-, Chondriosomen-, resp. Sphaerosomen-, (Mikrosomen)- und einer Zytoplasmavererbung. *Cytologia* **20**, 315–338.

Orias, E. & Flacks, M. (1975). Macronuclear genetics of *Tetrahymena*. I. Random distribution of macronuclear gene copies in *T. pyriformis*, syngen 1. *Genetics* **79**, 187–206.

Perlman, P. S. & Birky, C. W., Jr. (1974). Mitochondrial genetics in bakers' yeast: a molecular mechanism for recombinational polarity and suppressiveness. *Proceedings of the National Academy of Sciences, U.S.A.* **71**, 4612–4616.

Radding, C. (1973). Molecular mechanisms in genetic recombination. *Annual Review of Genetics* **7**, 87–111.

Rowlands, R. T. & Turner, G. (1974). Recombination between the extranuclear genes conferring oligomycin resistance and cold sensitivity in *Aspergillus nidulans*. *Molecular and General Genetics* **133**, 151–161.

Smith, G. P. (1974). Unequal crossover and the evolution of multigene families. *Cold Spring Harbor Symposia on Quantitative Biology* **38**, 507–513.

Sparrow, A. H., Price, H. J. & Underbrink, A. G. (1972). A survey of DNA content per cell and per chromosome of prokaryotic and eukaryotic organisms: some evolutionary considerations. *Evolution of Genetic Systems* (ed. H. H. Smith), pp. 451–493. New York: Gordon and Breach.

Tartof, K. D. (1974). Unequal mitotic sister chromatid exchange and disproportionate replication as mechanisms regulating ribosomal RNA gene redundancy. *Cold Spring Harbor Symposia on Quantitative Biology* **38**, 491–500.

TARTOF, K. D. (1975). Redundant genes. *Ann. Rev. Genet.* **9** (in the Press).

TAYLOR, J. H., WOODS, P. S. & HUGHES, W. L. (1957). The organization and duplication of chromosomes as revealed by autoradiographic studies using tritium labeled thymidine. *Proceedings of the National Academy of Sciences, U.S.A.* **43**, 122–128.

THOMAS, C. A., JR. (1970). The theory of the master gene. *The Neurosciences: Second Study Program* (ed. F. O. Schmitt), pp. 973–978. New York: Rockefeller University Press.

THOMAS, C. A., JR. (1974). The rolling helix: a model for the eukaryotic gene? *Cold Spring Harbor Symposia on Quantitative Biology* **38**, 347–352.

VISCONTI, N. & DELBRÜCK, M. (1953). The mechanism of genetic recombination in phage. *Genetics* **38**, 5–33.

WILLIAMSON, D. H. & FENNELL, D. J. (1974). Apparent dispersive replication of yeast mitochondrial DNA as revealed by density labelling experiments. *Molecular and General Genetics* **131**, 193–207.