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Genetics

# The frequency of meiotic recombination in yeast is independent of the number and position of homologous donor sequences: Implications for chromosome pairing

(ectopic recombination/*Saccharomyces cerevisiae*)

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**ABSTRACT** **We constructed diploids of *Saccharomyces cerevisiae* homozygous for *LEU2* and carrying one, two, or four copies of *leu2* at ectopic locations and determined the frequency of 3+:1− (*LEU2*:*leu2*) meiotic tetrads. Gene conversion between a *LEU2* recipient and a *leu2* ectopic donor occurred at the same frequency as did gene conversion between allelic copies of *LEU2* and *leu2*. An increase in the number of possible ectopic donor loci did not lead to a proportional increase in the level of ectopic gene conversion. We suggest that the limiting step in meiotic recombination is the activation of a locus to become a recipient in recombination and that once activated, a locus can search the entire genome for a homologous partner with which to recombine. In this respect, this search for a homologous partner resembles the efficient premeiotic methylation/inactivation of duplicated sequences in *Ascobolus* and *Neurospora*. These observations support models in which strand exchange serves to align homologous chromosomes prior to their becoming much more fully synapsed by the elaboration of the synaptonemal complex.**

The high level of homologous recombination in meiosis is generally thought to be dependent on the pairing and alignment of homologous chromosomes and the formation of the synaptonemal complex (SC) (reviewed in refs. 1–4). Certainly, there is a strong correlation between the appearance of the SC and the high frequency of meiotic recombination. In *Saccharomyces cerevisiae*, a number of null mutations that are defective in meiotic recombination also lack a normal SC (5–10). However, these mutations do not allow one to distinguish whether SC formation precedes meiotic recombination or, conversely, if DNA strand exchange between homologous chromosomes establishes the alignment of chromosomes that then become more fully synapsed by the formation of SC.

The view that the formation of SC is a necessary precondition for a high level of meiotic recombination is disturbed by recent observations of frequent ectopic recombination between short homologous sequences in nonallelic locations (11–13). In *S. cerevisiae*, ectopic meiotic gene conversions at frequencies as high as 1% of all meioses were first detected between artificially duplicated copies of the 2.2-kilobase (kb) *LEU2* gene (11, 13) and the 6-kb *HIS3* gene (12). Ectopic meiotic gene conversions and crossovers also occur frequently between members of naturally repeated gene families, including Ty elements (14–16) and the Y′ subtelomeric family (17). Similar events have also been observed in mitotic cells (16–18). In all respects, ectopic recombination resembles recombination between the same sequences at their normal, allelic position. Ectopic gene conversions of naturally and artificially repeated sequences are frequently associated with crossing-over (11–15, 17). The frequency with which particular alleles undergo gene conversion and mismatch repair is the same at ectopic and homologous sites (13, 19). Finally, the *red1* and *hop1* mutations that reduce (but do not eliminate) normal interhomologue recombination also reduce ectopic recombination (10, 20).

The observation of high levels of ectopic recombination raises several important questions about the way that homologous sequences "find each other" in order to undergo recombination and the rules that govern their interaction. We consider two general classes of models that might explain how recombining sequences interact. In one class of models, the rate-limiting step is the activation of a particular sequence to initiate recombination. Subsequent steps are not rate limiting; the activated region always succeeds in finding a homologous partner. This "obligate recombination" class includes both models in which an activated sequence could serve as a donor or as a recipient. In either case, the frequency with which a particular locus becomes a recipient in meiotic gene conversion will be, to a first approximation, independent of the number of possible donor loci. If gene conversion among a family of repeated sequences is initiated by activation of one of these sequences to act as a donor, it will have $n - 1$ possible partners in a set of $n$ repeated sequences, only one of which is the recipient of interest. Hence, the chance that a given donor will recombine with a designated recipient is only $1/(n - 1)$. This is true for each donor; hence the frequency that a designated recipient will be converted by all possible donors is no different from the case when there was only a single ectopic donor: $(n - 1) \times 1/(n - 1) = 1$. A similar conclusion prevails for models where gene conversions result from the activation of one sequence to act as a recipient. If an activated locus will invariably find a homologous, non-sister sequence with which to recombine, the frequency with which an ectopic interaction will occur will also be largely independent of the number of potential ectopic donors (15).

Another class of models assumes that the rate-limiting step involves the pairing and exchange of DNA strands between homologous partners—that is, the likelihood of an activated sequence locating and recombining with a homologous non-sister partner will be low. For example, homologous sequences might encounter each other randomly but only a small fraction of these encounters might then lead to an exchange of genetic information. Alternatively, activation of a locus to be a recipient in gene conversion may be transient, with the possibility of repairing a recombinogenic DNA lesion by sister-chromatid recombination or by religation of the lesion as well as by recombining with non-sister homol-

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Abbreviation: SC, synaptonemal complex.
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ogous sequences. In these models, the frequency with which a given recipient will undergo a detectable recombination event will increase proportionally to the number of possible homologous donors. Although they recognized several alternative interpretations Kupiec and Petes (15) favored this mechanism when they argued that a correction factor accounting for multiple copies of the Ty*1* retrotransposon be applied to their observed frequency of gene conversion between Ty*1* elements. This leads to an estimate of the frequency of Ty × Ty conversion that is 200-fold lower than the frequency of recombination between duplicated unique sequences.

To test whether the rate of ectopic recombination is dependent on the number of ectopic donors, we have constructed a series of isogenic diploids homozygous for the wild-type *LEU2* gene and carrying one, two, or four copies of the double mutant *leu2-A,R* allele at different ectopic locations. Meiotic gene conversion events in which one of the *LEU2* alleles is the recipient of mutant information yields a tetrad containing three Leu$^+$ and one Leu$^-$ spore. Our results show that the frequency of ectopic recombination is largely independent of the number of potential donors. These and other data lead us to suggest that once a region has been activated to be a recipient during gene conversion, it efficiently finds a homologous partner independent of the donor's allelic or ectopic chromosome location. Our results support suggestions (3, 8, 21) that recombination plays a central role in homologous chromosome pairing.

## MATERIALS AND METHODS

**Strains.** All strains were isogenic derivatives of two haploid parents, G167 (WYL114-7A) and G170 (WYL115-4D), closely related meiotic segregants of the highly inbred strains used in previous studies of ectopic recombination (13). The important features of the diploids used in this work are described in Tables 1 and 2. Strains were transformed with pBR322 plasmids carrying the 1.1-kb *Hin*dIII *URA3* fragment at the *Hin*dIII site, the 2.2-kb *Sal* I–*Xho* I fragment of *leu2* at the *Sal* I site, and either the 3.5-kb *Hin*dIII fragment of *MAT***a** or the 1.3-kb *Pvu* II–*Cla* I fragment of *HIS4* (13). These plasmids were integrated at *MAT* or *HIS4*, respectively, as described (13). In each case, the plasmid's *leu2* region was modified to carry two 4-base-pair (bp) insertions, separated by 400 bp, at the *Eco*RI and *Asp*718 sites. The *leu2-R* mutation is an enzymatic fill-in of the *Eco*RI site in *LEU2*, creating a 4-bp duplication (13). The *leu2-A* mutation is allelic to the previously described *leu2-K* (*Kpn* I) mutation previously employed (12, 16), except that the *Kpn* I isoschizomer, *Asp*718, was used to create 5′ overhangs that were filled in to give another 4-bp duplication instead of the 4-bp deletion in *leu2-K*. The resulting double mutant is designated *leu2-A,R*. The use of the *leu2-A,R* double mutant increases substantially the probability that at least one of the two mutant sites will convert *LEU2*; ≈50% of the Leu$^-$ convertants are *leu2-A,R*, whereas the remainder are either *leu2-A* or *leu2-R* (data not shown). In all cases the *leu2-A,R* sequences were oriented in the same direction as *LEU2*.

Diploids WYL121, -122, -125, -127, -128, and -129 are isogenic, except for the integrative transformation of the *leu2-A,R* sequences (Table 1). Strains WYL131 and -130 were obtained by crossing *his4::(URA3-leu2-A,R) MAT::(URA3-leu2-A,R)* segregants of diploids WYL126 and -127. Given the highly backcrossed origin of the parent haploid strains (13), diploids WYL130 and -131 are very closely related to the other strains used in the study. The two haploid parents of WYL140 were obtained by selecting His$^+$ Ura$^-$ derivatives of the two haploid parents of WYL139 on medium lacking

Table 1. Strains

| | |
|---|---|
| Haploid | |
| G170 | *MAT***a** *ura3-1 trp5 met13-2 ade1 lys2 can1 cyh2 LEU2* |
| G167 | *MATα ura3 trp1 met13-2 ade1 lys2 LEU2* |
| G176 | G170 except [*his4′*-pBR322-*URA3-leu2-A,R-′his4*] |
| G173 | G167 except [*his4′*-pBR322-*URA3-leu2-A,R-′his4*] |
| G184 | G170 except [*MAT***a**-pBR322-*URA3-leu2-A,R-MAT***a**] |
| G183 | G167 except [*MATα*-pBR322-*URA3-leu2-A,R-MATα*] |
| G187 | [*MAT***a**-pBR322-*URA3-leu2-A,R-MAT***a**][*his4′*-pBR322-*URA3-leu2-A,R-′his4*] *ura3-1 trp1 met13-2 ade1 lys2 cyh2 LEU2* |
| G189 | [*MATα*-pBR322-*URA3-leu2-A,R-MATα*][*his4′*-pBR322-*URA3-leu2-A,R-′his4*] *ura3-1 trp5 met13-2 ade1 lys2 can1 cyh2 LEU2* |
| G188 | [*MAT***a**-pBR322-*URA3-leu2-A,R-MAT***a**][*his4′*-pBR322-*URA3-leu2-A,R-′his4*] *ura3-1 trp5 met13-2 ade1 lys2 can1 cyh2 LEU2* |
| G190 | [*MATα*-pBR322-*URA3-leu2-A,R-MATα*][*his4′*-pBR322-*URA3-leu2-A,R-′his4*] *ura3-1 trp1 met13-2 ade1 lys2 cyh2 LEU2* |
| G191 | *MAT***a** *leu2-A,R* [*his4′*-pBR322-*URA3-leu2-A,R-′his4*] *ura3-1 trp1 met13-2ade1 lys2 can1 cyh2* |
| G192 (G173 His$^+$) | *MATα ura3-1 trp1 met13-2 ade1 lys2 cyh2* |
| G193 (G191 His$^+$) | *MAT***a** *leu2-A,R ura3-1 trp1 met13-2 ade1 lys2 can1 cyh2* |
| Diploid* | |
| WYL121 | G173 × G176 |
| WYL122 | G173 × G170 |
| WYL125 | G183 × G184 |
| WYL126 | G173 × G184 |
| WYL127 | G183 × G176 |
| WYL128 | G183 × G170 |
| WYL128A | G183 × G170 |
| WYL129 | G167 × G184 |
| WYL130 | G187 × G189 |
| WYL131 | G188 × G190 |
| WYL139 | G191 × G173 |
| WYL140 | G193 × G192 |

*The arrangement of ectopic *leu2-A,R* sequences in each diploid is shown in Tables 2 and 3.

histidine. Thus, WYL139 and -140 are isogenic, except that WYL140 lacks the *leu2-A,R* sequences inserted at *HIS4*.

**Genetic Methods.** Diploids were sporulated, dissected, and analyzed as described (13). The nature of the *LEU2* to *leu2* conversion event (to *leu2-A*, *leu2-R*, or *leu2-A,R*) was determined by allele testing the *leu2* segregant in each tetrad as described (13). Crossing-over between *leu2-A,R* inserted at *HIS4* and sequences at *LEU2* yields a nonlethal deletion of ≈20 kb, including the 5′ end of *HIS4*. These crossover products are detected as colonies that are unable to papillate to $His^+$ (13).

## RESULTS

**Gene Conversion of *LEU2* by Ectopic *leu2-A,R* Donors.** A series of *LEU2/LEU2* diploids were constructed containing one, two, or four copies of *leu2-A,R* sequences inserted either at *HIS4* (20-kb centromere distal to *LEU2*) or *MAT* (70 kb from *LEU2*, on the opposite side of the centromere) (Fig. 1 and Table 2). Approximately 220 tetrads from each strain were dissected and scored for gene conversions of *LEU2* to *leu2* [which can be to either *leu2-A*, *leu2-R*, or *leu2-A,R*, depending on whether one or both markers are converted (ref. 12; see also *Materials and Methods*)]. The results of this analysis are shown in Table 2 and Fig. 1. Meiotic conversion of *LEU2* to *leu2* occurred in 3–7% of the tetrads.

The ectopic copies of *leu2-A,R* inserted at *MAT* and at *HIS4* acted as donors with similar efficiencies. 3+:1− segregation was observed in 3.3% of tetrads from diploids with a single copy of *leu2-A,R* at *MAT* (WYL128 and -129) and in 4.7% of tetrads from diploids with a single copy of *leu2-A,R* inserted at *HIS4* (WYL122). The reduction in ectopic conversions recovered from diploids with *leu2-A,R* inserted at *MAT* is expected, because gene conversions accompanied by crossing-over between *MAT::leu2-A,R* and *LEU2* generate lethal deficiency or dicentric chromosomes (11, 13). In contrast, conversions with and without crossing-over between *his4::leu2-A,R* and *LEU2* are both viable (13). Indeed, approximately half (15/26) of the 3+:1− tetrads from diploids WYL121 and -122 contained a spore with a deletion between *his4::leu2-A,R* and *LEU2* (see *Materials and Methods*).

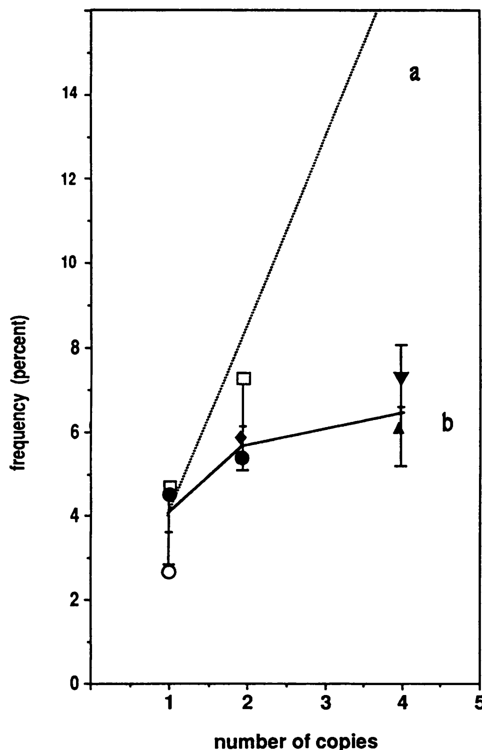


FIG. 1. Frequency of ectopic meiotic gene conversion of *LEU2* by one or more *leu2-A,R* donor sequences. Data are from Table 1. Calculated standard deviations for each set of data are indicated by vertical lines. Line a is that expected if the frequency of gene conversion events is proportional to the number of possible donors in each diploid, based on the observed average frequency for one donor. Line b is that expected if the total frequency of gene conversion events for a *LEU2* recipient is a constant (assumed here to be 8%) and where either *LEU2* (allelic) or *leu2-A,R* (ectopic) sequences are equally likely to act as donors (see text). Standard deviations are indicated by lines with small bars. ○, WYL128; □, WYL122 or WYL121; ●, WYL129 or WYL125; ◆, WYL127; ▼, WYL130; ▲, WYL131.

Table 2. Frequency of ectopic *LEU2* gene conversion (3+:1−) among tetrads

| Diploid genotype | Strain* | Conversion | Frequency, % |
|---|---|---|---|
| *his4::leu2-A,R LEU2 MAT* / *HIS4 LEU2 MAT* | WYL122 | 10/211 | 4.7 |
| *HIS4 LEU2 MAT::leu2-A,R* / *HIS4 LEU2 MAT* | WYL128 | 13/492 | 2.6 |
| | WYL129 | 11/243 | 4.5 |
| *his4::leu2-A,R LEU2 MAT* / *his4::leu2-A,R LEU2 MAT* | WYL121 | 16/218 | 7.3 |
| *HIS4 LEU2 MAT::leu2-A,R* / *HIS4 LEU2 MAT::leu2-A,R* | WYL125 | 12/221 | 5.4 |
| *his4::leu2-A,R LEU2 MAT* / *HIS4 LEU2 MAT::leu2-A,R* | WYL127 | 19/319 | 5.9 |
| *his4::leu2-A,R LEU2 MAT::leu2-A,R* / *his4::leu2-A,R LEU2 MAT::leu2-A,R* | WYL130 | 19/261 | 7.3 |
| | WYL131 | 9/150 | 6.0 |

*All diploids are isogenic except WYL130 and -131, which are constructed from segregants of strain WYL126 and -127.

**Ectopic Gene Conversions Do Not Increase in Proportion to the Number of Donors.** The data presented in Table 2 are not consistent with models in which the frequency of ectopic recombination increases in direct proportion to the number of potential ectopic donors (illustrated by line a in Fig. 1). Given an average frequency of gene conversion when there is one ectopic donor of 3.6% (WYL122, -128, and -129), this model would predict 14.4% of tetrads from diploids with four potential donors to display 3+:1− segregation. In fact, only 6.8% of tetrads from these strains (WYL130 and -131) displayed ectopic gene conversion, a frequency significantly less than the 14.4% predicted [$P < 0.001$ based on a G test (22)].

Another way to demonstrate that the frequency of ectopic gene conversions is not a linear function of the number of donors is to compare the frequency of 3+:1− tetrads for diploids WYL130 and -131 (with two donor copies at *HIS4* and two more at *MAT*) with the sum of the frequencies of diploids WYL121 (two donors at *HIS4*) and WYL125 (two donors at *MAT*). This comparison should correct for the fact that some of the ectopic recombinations between *LEU2* and *MAT::leu2-A,R* will produce inviable products (13). The combined frequency of 3+:1− tetrads in the latter two strains (12.7%) is significantly greater ($P < 0.05$) than the observed frequency for strains WYL130 and -131 (6.8%).

**Allelic and Ectopic Events Occur at Equivalent Frequencies but Are Not Additive.** To demonstrate that ectopic events occur at frequencies comparable to allelic events, we measured the number of *LEU2* to *leu2* gene conversions in a diploid of genotype *LEU2/leu2-A,R* (WYL140, Table 3). The frequency of conversion events in this strain, where all of the

Table 3. Frequency of allelic and/or ectopic *LEU2* gene conversion among tetrads

| Strain | Diploid genotype | 3Leu$^-$:1Leu$^+$ conversion | Frequency, % |
|---|---|---|---|
| WYL140 | *HIS4* *LEU2* *MAT* / *HIS4* *leu2-A,R* *MAT* | 7/271 | 2.6 |
| WYL139 | *his4::leu2-A,R* *LEU2* *MAT* / *his4::leu2-A,R* *leu2-A,R* *MAT* | 9/279 | 3.2 |

conversions result from interactions between sequences at allelic positons, is 2.8%. This value is approximately the same as that found for diploid WYL122 (Table 2), once the frequency of ectopic gene conversion is corrected to reflect the presence of two possible *LEU2* recipients (2.3%). In strain WYL122, all 3+:1− tetrads are produced by ectopic conversions between *leu2-A,R* and *LEU2*.

In addition, we have compared the frequency of *LEU2* to *leu2* gene conversions in diploid WYL140 (*LEU2/leu2-A,R*) with an isogenic strain (WYL139) that has two ectopic copies of *leu2-A,R* at *HIS4* in addition to the *leu2-A,R* allele at its normal location (Table 3). The frequencies of 3−:1+ tetrads in these two diploids are statistically indistinguishable. Hence, allelic gene conversions do not occur at a significantly higher frequency than ectopic events, and the total number of events is not augmented by additional (ectopic) copies.

## DISCUSSION

**Equal Frequencies of Allelic and Ectopic Gene Conversions Suggest a Global Search for Homologous Sequences.** The data presented here and in previous studies (12, 13) can be summarized in three statements. (*i*) The frequency of ectopic gene conversion is comparable to the allelic frequency. (*ii*) The frequency of ectopic events does not increase proportionally to the number of ectopic donors. (*iii*) The total number of ectopic gene and allelic gene conversions involving a given recipient sequence is not additive but rather is approximately constant.

These results are consistent with a mechanism for meiotic recombination in which one of the *LEU2* genes on the four chromatids is activated (by nicking or cutting) in any given meiosis. Once activated, a sequence will be the recipient of a genetic exchange event. The activated *LEU2* locus will pair and recombine with any available homologous partner, irrespective of the genomic location of that partner. Therefore, in a *LEU2/LEU2* diploid with one ectopic *leu2-A,R* donor, 50% of the interactions of an activated *LEU2* sequence will be with its allelic *LEU2* partner and 50% will be with the ectopic *leu2-A,R* donor; only the latter will yield 3+:1− gene conversions. With two ectopic donors, the proportion of interactions between the recipient and the ectopic *leu2-A,R* alleles increases to 67%; with four donors, it increases to 80%. (Repair by way of an interaction with a sister chromatid does not enter into this calculation, as such events simply reduce the number of times an activated gene could be detected as a recipient. These predictions are presented graphically in Fig. 1, line b.) A good fit of the experimental data is obtained by assuming that the *LEU2* locus is activated in about 8% of meioses. We conclude that the predominant rate-limiting step in meiotic recombination is the activation of a recipient. If both initiation and pairing/exchange were equivalently rate-limiting steps, the frequency of gene conversion would also be expected to increase linearly with the number of potential donors.

We have assumed that the rate-limiting step in meiotic recombination is the activation of a locus to become a recipient. As we pointed out in the Introduction, the data could equally be accommodated by models in which the activated sequence became a donor of information. However, previous studies have provided strong evidence that recombinationally active loci are, in fact, preferential recipients of genetic information (13, 23, 24). For example, the same pair of *leu2* heteroalleles, moved to five different chromosomal locations, exhibits a 20- to 40-fold difference in the frequency of allelic gene conversions (13). When ectopic gene conversions between "hot" and "cold" loci are examined, nearly all of the conversion events are at the "hotter" of the two loci (13). Further support is provided by Lambie and Roeder's (23) finding that the insertion of a centromere near a locus decreased the frequency with which that locus received information during a gene conversion. Finally, the analysis of Nicolas *et al.* (24) of the effects of deleting a meiotic "hot spot" near the yeast *ARG4* gene also indicates that a recombinationally active locus is usually the recipient.

The equivalence of allelic or ectopic copies as the donor implies that interactions of recombining homologous sequences in yeast are essentially random, independent of chromosome location. In this paper we have only examined ectopic recombination between loci >20 kb apart on the same chromosome, but other work has shown that ectopic recombination occurs at similar frequencies between sequences on nonhomologous chromosomes (12, 13, 17, E. J. Louis and J.E.H., unpublished). One study that showed much lower ectopic recombination than allelic recombination involved a marked Ty element (16) and may have been biased by the fact that the *URA3* marker was inserted only 300 bp from the end of the repeated sequence. The limited amount of homology on one side of the marker that was to be deleted by gene conversion is the 300-bp long terminal repeat $\delta$ sequence of Ty, which also exhibits more DNA sequence variation than most of the Ty element. It is, of course, possible that some variation exists in the accessibility of some ectopic locations to serve as donors during ectopic interactions; for example, repeated sequences that are very close together (much less than 20 kb apart) might act as preferred substrates. For simplicity, we have assumed that all ectopic sites are similar in their ability to act as donors.

The observation that a recipient is as likely to recombine with a homologous ectopic sequence as with an allelic copy on its opposite homologue suggests that the extent of pairing of homologous regions during recombination occurs over relatively short distances. The extent of homology shared by *LEU2* and the ectopic *leu2-A,R* is only 2.2 kb, whereas the shared homology between the *LEU2* alleles on opposite homologues is effectively infinite. Consequently, 2 kb of homology is sufficient for an activated locus to recognize efficiently a homologous sequence. This length of DNA is similar to the average meiotic gene conversion tract length (1–2 kb) measured at several loci (25–27).

In an earlier study of ectopic recombination involving a comparison of the meiotic ectopic recombination between moderately repeated Ty*1* transposon sequences and a duplication of *URA3* sequences, Kupiec and Petes (15) found that the observed rates were quite similar but argued that the intrinsic rate for Ty*1* × Ty*1* interactions was actually much lower, because of the many copies of Ty*1* in the genome. Our data suggest that a correction based on the number of possible donor sequences is not warranted, leading to the conclusion that there has been no evolutionary selection for a reduced propensity of naturally repeated sequences to undergo meiotic ectopic recombination. This conclusion is further supported by our recent study of frequent ectopic recombination between the repeated Y′ sequences at many chromosome ends (ref. 17; E. J. Louis and J.E.H., unpublished observations).

**A Recipient May Search the Entire Genome for a Homologous Partner.** The suggestion that yeast cells in meiosis have

the capacity to search the entire genome for homologous partners is not without precedent in other organisms. In *Neurospora* (28, 29) and *Ascobolus* (30) duplicated genes at different chromosomal locations are inactivated by methylation during the heterokaryon stage that follows conjugation and precedes meiosis. A pair of duplicated sequences has better than a 50% chance of being methylated (29, 30). The "homology sensing" machinery of these filamentous fungi and of *Saccharomyces* may be analogous, except that the yeast machinery would be associated with (or is) a recombination enzyme complex and the *Neurospora* and *Ascobolus* enzymes would be associated with a methylase.

**Relationship Between Chromosome Pairing and Recombination.** If activated sequences can find a homologous partner by searching the genome for homology, what then is the origin and role of the synaptonemal complex? The fact that ectopic recombination is as frequent as allelic events makes it unlikely that the primary role of the SC is to align long regions of homologous chromosomes to enhance the frequency of strand exchange. We believe the SC plays other central roles in the way in which strand exchange events are resolved. For example, SC may prevent or limit the number of gene conversions that are resolved as *bona fide* crossovers, between sister chromatids and between non-sister chromatids. The SC may also ensure proper chromosomal disjunction after exchanges are completed. It is also important to note that only a small fraction of chromosomal DNA may actually be embraced by SC, so that much of the DNA would still be free to interact with homologous partners independent of chromosome position. However, the strong correlation between the number and distribution of recombination nodules along SC and the distribution of crossover events (3) argues that that DNA undergoing recombination is closely associated with the SC.

This intimate relationship between genetic exchange and the formation of SC is preserved if one assumes that it is indeed the initiation of exchange that establishes the pairing of chromosomes. The isolation of mutations that affect chromosome pairing and recombination should be valuable in testing this idea. A model that assumes recombination is a prerequisite for chromosome pairing predicts that all mutations that significantly impair recombination will perforce disrupt the synapsis of homologous chromosomes. This need not be the case if, instead, chromosome pairing proceeds independent of actual strand exchange. To date, there are no mutations that either (*i*) exhibit nearly normal recombination but do not form normal SC or (*ii*) have very little recombination but nearly normal SC. Two mutations are, however, informative. Both *red1* (10) and *hop1* (6, 7) mutants, which appeared primarily to affect homologous chromosome pairing and the formation of SC, now have been shown to reduce both allelic and ectopic recombination to the same extent (10, 20). These phenotypes are consistent with models in which recombination precedes chromosome pairing.

In our view, the pairing and synapsis that occurs between homologous chromosomes in yeast is a reflection of the fact that most DNA along a chromosome is single-copy and that the activation of a sequence to initiate a search for homology will only yield its properly aligned allelic partner. In higher eukaryotes, this same mechanism of chromosome pairing may also apply, despite the fact that there is substantially more repeated DNA. Much of the repeated DNA is in heterochromatic regions that may not be activated for recombination or else may be too short to find homologous ectopic sequences efficiently. There may, in addition, be a few preferential pairing sites, reflecting other mechanisms of preliminary alignment that bias interactions toward homologues; indeed, in *Caenorhabditis* (31, 32) and in *Drosophila* (33, 34) there is genetic evidence of important pairing sites that permit adjacent regions of chromosomes to recombine. Our data suggest that the predominant way in which homologous sequences interact during recombination in yeast is largely independent of chromosomal position.

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