Enhancement of spontaneous mitotic recombination by the meiotic mutant *spoil-1* in *Saccharomyces cerevisiae*

(crossing-over/gene conversion/mitosis)

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ABSTRACT Both nonreciprocal and reciprocal mitotic recombination are enhanced by the recessive mutant *spoil-1*, which was previously shown to affect meiosis by decreasing recombination and increasing nondisjunction. The mitotic effects are not distributed equally in all chromosomal regions. The genotypes of mitotic recombinants in *spoil-1/spoil-1* diploid cells provide further evidence that widely spaced chromosomal markers undergo coincident conversion in mitosis.

Diploid cells homozygous for the recessive mutation *spoil-1* exhibit diminished sporulation, $\approx 0.02\%$ ascospore survival at 24–34°C, a meiotic Rec[−] phenotype with respect to both intergenic recombination and intragenic recombination resulting in prototrophy, and high levels of meiotic chromosomal nondisjunction (1–3). In this study, we examined the effects of the *spoil-1* mutation on mitotic intragenic and intergenic recombination. Diploid *spoil-1/spoil-1* hybrids are proficient in both nonreciprocal intragenic and intergenic recombination and reciprocal intergenic recombination. The genotypes of sectorial colonies indicate, however, that the *spoil-1* mutation increases the length of chromosomal regions exhibiting coincident gene conversion and thereby alters the disposition of reciprocal exchanges. The properties of mitotic recombination in *spoil-1/spoil-1* cells are consistent with the prereplicative model of mitotic recombination (4) and the Aviemore model of genetic recombination, in which Holliday structures resulting in reciprocal events are established by isomerization following asymmetric transfer of single-stranded DNA from a donor to a recipient chromosome (5). Several of the effects of the *spoil-1* mutation in meiosis and mitosis are explicable in terms of the altered recombinational phenotype displayed by *spoil-1/spoil-1* diploids.

MATERIALS AND METHODS

Yeast Strains. The genotypes of the diploid hybrids used and the linkage relations of chromosome 7 markers relevant to the experiments described are given in Table 1. The *spoil-1/spoil-1* and *SPO11/SPO11* hybrids are coisogenic strains closely related in genetic background. Each parent of the *spoil-1/spoil-1* hybrid resulted from at least five back-crosses to the *SPO11* strains used. The meiotic map order of the *leu1-c*, *leu1-12*, *trp5-c*, and *trp5-d* heteroalleles was determined by standard non-selective tetrad analysis (6).

Media. The recipes for growth and sporulation media have been reported (7).

Rates of Mitotic Recombination. The rates of spontaneous mitotic recombination at 24°C resulting in tryptophan proto-

Table 1. Strains used

Diploid	Relevant genotypes					
CBX1	<i>spoil-1</i>	<i>MATα</i>	<i>ade2-1</i>	<i>can1^r</i>		
	<i>spoil-1</i>	<i>MATa</i>	<i>ade2-1</i>	<i>CAN1^s</i>		
	<i>ADE5</i>	<i>met13-c</i>	<i>cyh2^r</i>	<i>trp5-c</i>	<i>leu1-c</i>	<i>ade6</i>
	<i>ade5</i>	<i>MET13</i>	<i>CYH2^s</i>	<i>trp5-d</i>	<i>LEU1</i>	<i>ADE6</i>
CBX2	<i>SPO11</i>	<i>MATα</i>	<i>ade2-1</i>	<i>can1^r</i>		
	<i>SPO11</i>	<i>MATa</i>	<i>ade2-1</i>	<i>CAN1^s</i>		
	<i>ADE5</i>	<i>met13-c</i>	<i>cyh2^r</i>	<i>trp5-c</i>	<i>leu1-c</i>	<i>ADE6</i>
	<i>ade5</i>	<i>MET13</i>	<i>CYH2^s</i>	<i>trp5-d</i>	<i>LEU1</i>	<i>ADE6</i>
JG44	<i>SPO11</i>	<i>MATα</i>	<i>ade2-1</i>			
	<i>SPO11</i>	<i>MATa</i>	<i>ade2-1</i>			
	<i>ADE5</i>	<i>met13-d</i>	<i>CYH2^s</i>	<i>trp5-d</i>	<i>leu1-12</i>	<i>ADE6</i>
	<i>ade5</i>	<i>met13-c</i>	<i>cyh2^r</i>	<i>trp5-c</i>	<i>leu1-c</i>	<i>ADE6</i>
JG61	<i>SPO11</i>	<i>MATα</i>	<i>ade2-1</i>			
	<i>SPO11</i>	<i>MATa</i>	<i>ade2-1</i>			
	<i>ADE5</i>	<i>met13-d</i>	<i>CYH2^s</i>	<i>trp5-d</i>	<i>leu1-12</i>	<i>ADE6</i>
	<i>ade5</i>	<i>met13-c</i>	<i>cyh2^r</i>	<i>trp5-c</i>	<i>leu1-c</i>	<i>ADE6</i>

Meiotic linkage map of chromosome 7 (centimorgans)

trophs due to intragenic recombination and segregation of the recessive drug-resistance markers *can1^r* and *cyh2^r* were estimated by the method of the median of Lea and Coulson (8) as described (4, 6, 9).

RESULTS

Spontaneous Mitotic Recombination in Coisogenic *spoil-1/spoil-1* and *SPO11/SPO11* Hybrids. The *spoil-1/spoil-1* hybrid CBX1 and the *SPO11/SPO11* hybrids CBX2, JG44, and JG61 were used to assess the effect of the *spoil-1* mutation on mitotic recombination. Intragenic recombination at the *TRP5* locus, resulting in Trp⁺ prototrophs, monitored intragenic events on chromosome 7. The genotypes of the hybrids also allow detection of intergenic recombination associated with conversional events at the *TRP5* locus. The diploids are *ade2-1/ade2-1 ADE5/ade5* hybrids and form red colonies because of the cell-limited pigment accumulated by *ade2-1* mutants (10). Intergenic recombination, associated with intragenic recombination

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at *TRP5*, yields *ade5/ade5* Trp⁺ segregants. These cells form white colonies because the *ade5* mutation blocks accumulation of the red pigment precursor (10). Red-white-sectored Trp⁺ colonies are also detected when conversion at the *TRP5* locus yields a pair of prototrophic cells during the cell division preceding or immediately after plating on tryptophanless medium (4, 6, 11). The ensemble of mitotic events that results in red-white sectoring was monitored by the appearance of sectored colonies on synthetic complete medium. Mitotic segregation of the recessive drug-resistance marker *cyh2*^r, located on chromosome 7, and *can1*^r, located on chromosome 5, provided a further basis for comparison.

Mitotic events that result in total tryptophan prototrophic intragenic recombinants, cycloheximide-resistant segregants, and canavanine-resistant segregants occur at similar rates in *spo11-1/spo11-1* and *SPO11/SPO11* hybrids (Table 2). Sectored colonies resulting from segregation of the *ade5* mutation, and markers linked to it, recovered on synthetic complete medium, are four times more frequent in the *spo11-1/spo11-1* hybrid. The enhancement is highly statistically significant (Table 2).

Genotypic Analysis of Red-White-Sectored Colonies of *spo11-1/spo11-1* and *SPO11/SPO11* Hybrids. The genotypes of sectored Trp⁺ colonies and sectored colonies recovered on synthetic complete medium were determined to further characterize the effects of the *spo11-1* mutation in mitotic cells. In *SPO11/SPO11* cells, the two types of sectors reflect different classes of recombinational events. Approximately 80% of prototrophic red-white-sectored colonies result from associated reciprocal recombination in the vicinity of the site of intragenic recombination. The remainder exhibit nonreciprocal intergenic recombination, which can extend from the site of intragenic recombination to the *ADE5* locus (4, 6, 11). In contrast to sectored prototrophic colonies, ≈80% of red-white-sectored colonies of spontaneous origin, recovered nonselectively on synthetic complete medium, result from nonreciprocal segregation that may involve the signaling *ade* marker as well as markers linked to it (12–14).

Red-White Sectors Recovered on Synthetic Complete Medium. We determined the genotypes of 51 red-white sectors recovered from platings of the *spo11-1/spo11-1* hybrid on synthetic complete medium to determine whether the enhanced sectoring is a result of stimulation of nonreciprocal or reciprocal

recombination (or both) (Table 3). The *spo11-1* mutation increases the frequency of both reciprocal and nonreciprocal events resulting in mitotic segregation; 43/51 (84%) of the sectors exhibited only nonreciprocal sectoring for chromosome 7 markers, 8/51 (16%) sectors (classes 4, 5, 8, and 10) exhibited reciprocal segregation at the *ADE5* locus and three of these (classes 5, 8, and 10) were accompanied by nonreciprocal events at the *CYH2* and *MET13* loci. The presence of heterozygous markers at *LEU1* and *ADE6* on opposite sides of the centromere of chromosome 7 provided an opportunity to determine whether nonreciprocal intergenic recombination events can cross a centromere. We observed no instances of coincident conversion involving the *ADE6* and *LEU1* loci among a total of 14 sectors (classes 1, 2, 16, and 17) in which nonreciprocal segregation occurred at either *ADE6* or *LEU1*. The genotypes of 6 red-white-sectored colonies of the *SPO11/SPO11* hybrid JG61 were also determined (Table 4); 5/6 result from nonreciprocal events (classes 1 and 2) and 1/6 results from reciprocal recombination in the *MET13-ADE5* interval (class 3).

Red-White-Sectored Trp⁺ Colonies. We determined the genotypes of 17 Trp⁺ red-white-sectored colonies recovered from platings of the *spo11-1/spo11-1* diploid on tryptophan-free medium (Table 5) to inquire whether sectoring resulted from reciprocal intergenic recombination associated with intragenic recombination at the *TRP5* locus. In the *SPO11/SPO11* hybrids JG44 and JG61, 71 red-white-sectored Trp⁺ colonies have been described (4, 6, 11); 60/71 (85%) exhibited reciprocal intergenic recombination in the interval *TRP5-CYH2*, resulting in reciprocal segregation of markers at the *CYH2*, *MET13*, and *ADE5* loci. The *spo11-1/spo11-1* red-white-sectored Trp⁺ colonies exhibit a different pattern of segregation of markers at these loci. None of the *spo11-1/spo11-1* Trp⁺-sectored colonies reflect simple reciprocal intergenic recombination in the interval *TRP5-CYH2* and concomitant reciprocal segregation of markers at the distal *CYH2*, *MET13*, and *ADE5* loci (Table 5); 12/17 (71%) of the sectors (classes 1, 2, 4, 8, 9, and 10) result from conversional events involving the *CYH2*, *MET13*, and *ADE5* loci and 5/17 (classes 3, 5, 6, and 7) exhibit reciprocal segregation at the *ADE5* locus because of apparent reciprocal intergenic recombination in the most centromere distal interval, *MET13-ADE5*.

DISCUSSION

The data presented above show that the *spo11-1* mutant alters the properties of mitotic recombination in three respects: (i) it results in a nonuniform enhancement of reciprocal and nonreciprocal recombination; (ii) it enhances the relative proportion of intragenic conversional events at the *TRP5* locus, associated with coincident conversion of widely separated markers distal to the *TRP5* locus; and (iii) it abolishes the occurrence of intragenic conversional events at the *TRP5* locus, associated with simple intergenic recombination in the *TRP5-CYH2* interval, unaccompanied by conversion at the *CYH2*, *MET13*, and *ADE5* loci. These effects of the *spo11-1* mutation provide evidence that the *SPO11* gene product controls the length of chromosomal regions that undergo simultaneous gene conversion and concomitantly modulates the distribution of reciprocal intergenic exchanges.

The length of DNA segments that can exhibit coincident conversion of linked genetic markers and the position of associated reciprocal exchange events are mutually dependent properties of recombination according to the Aviemore model of recombination (5) and the prereplicative model of mitotic recombination in which recombination between homologous

Table 2. Mitotic rates of spontaneous intragenic recombination at the *TRP5* locus and segregation of heterozygous markers after growth of *spo11-1/spo11-1* and *SPO11/SPO11* hybrids

Recombinant class	Spontaneous mitotic rate	
	<i>spo11-1/spo11-1</i> (CBX1)	<i>SPO11/SPO11</i> (CBX2)
Trp ⁺ red	$1.63 \pm 0.17 \times 10^{-6}$	$1.71 \pm 0.25 \times 10^{-6}$
Trp ⁺ white	$4.07 \pm 0.49 \times 10^{-7}$	$2.72 \pm 0.47 \times 10^{-7}$
Trp ⁺ red-white sectored	1.90×10^{-7}	1.20×10^{-7}
Trp ⁺ total	$3.20 \pm 0.32 \times 10^{-6}$	$1.92 \pm 0.27 \times 10^{-6}$
<i>can</i> ^r	$2.38 \pm 0.20 \times 10^{-5}$	$3.39 \pm 0.38 \times 10^{-5}$
<i>cyh</i> ^r	$1.83 \pm 0.36 \times 10^{-5}$	$1.91 \pm 0.39 \times 10^{-5}$
Red-white sectored (<i>ade5</i>)*	$5.2 \pm 1.2 \times 10^{-4}$ (19/36,821)	$1.4 \pm 0.6 \times 10^{-4}$ (5/34,808)

*The frequency of red-white-sectored colonies (on synthetic complete medium) due to segregation of the *ade5* mutation in CBX1 is statistically significantly different than that observed in CBX2 ($\chi^2 = 6.9$; $N = 1$; $P < 0.01$).

Table 3. Genotypes of red-white-sectored colonies of the *spo11-1/spo11-1* hybrid CBX1 recovered on synthetic complete medium

Class	Genotypes observed												No. observed
	Red side						White side						
	ADE6	LEU1	TRP5*	CYH2	MET13	ADE5	ADE6	LEU1	TRP5*	CYH2	MET13	ADE5	
1	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{-}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	11
2	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{+}$	$\frac{-}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{+}$	1
3	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{-}$	13
4	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{-}$	5
5	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{-}$	1
6	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{-}$	1
7	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{-}$	3
8	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{-}$	1
9	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	6
10	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1
11	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1
12	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1
13	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{1+}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1
14	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{+2}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	2
15	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{1+}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{1+}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1
16	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{1+}{1+}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1
17	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{1+}{1+}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1

*1+ = *trp5-d*, and +2 = *trp5-c*.

chromosomes is initiated before completion of the mitotic DNA synthesis phase (4, 6, 11). The relevant events envisaged on the prereplicative model are shown in Fig. 1. Strand transfer establishes a region of heteroduplex DNA in the recipient DNA molecule only—i.e., asymmetric heteroduplex DNA. Strand

transfer is terminated by the isomerization postulated by Meselson and Radding (5) resulting in formation of a Holliday structure. Resolution of the Holliday structure by DNA replication during the mitotic DNA synthesis phase results in reciprocal recombination of DNA sequences flanking the region

Table 4. Genotypes of red-white-sectored colonies of the *SPO11/SPO11* hybrid JG61 recovered on synthetic complete medium

Genotypes observed*												No. observed
Class	Red side					White side						
	LEU1	TRP5	CYH2	MET13	ADE5	LEU1	TRP5	CYH2	MET13	ADE5		
1	+2	1+	—	+2	+	+2	1+	+	1+	—	4	
	1+	+2	+	1+	—	1+	+2	+	1+	—		
2	+2	1+	—	+2	+	+2	1+	—	+2	—	1	
	1+	+2	+	1+	—	1+	+2	+	1+	—		
3	+2	1+	—	+2	+	+2	1+	—	+2	—	1	
	1+	+2	+	1+	+	1+	+2	+	1+	—		

* At *LEU1*, 1+ = *leu1-c* and +2 = *leu1-12*; at *TRP5*, 1+ = *trp5-d* and +2 = *trp5-c*; at *MET13*, 1+ = *met13-c* and +2 = *met13-d*.

of asymmetric heteroduplex DNA and the Holliday junction (Fig. 1E). Genetic markers included in the region of asymmetric heteroduplex DNA will exhibit coincident gene conversion preserving their original coupling relations as a consequence of the replication of heteroduplex DNA (Fig. 1E). Coincident conversion of heterozygous markers in this region is not dependent on mismatch repair of heteroduplex DNA. If mismatch repair does occur, it can result in total elimination of one of the input alleles at a heterozygous site. This results in sectors in which both portions are homozygous for the same

allele of a locus previously in heterozygous condition (e.g., conversion at the *MET13* locus, classes 2, 5, 6, 11, and 17 of Table 3).

The precise nature of the *SPO11* gene product is not specified by the genetic results but the data favor the hypothesis that it is the postulated Meselson-Radding isomerase or a protein whose activity affects this type of isomerization. Delayed termination of asymmetric heteroduplex DNA formation by isomerization would be expected to result in the mitotic phenotype exhibited by the *spo11-1/spo11-1* diploid: diminished

Table 5. Genotypes of red-white-sectored Trp⁺ colonies of the *spo11-1/spo11-1* hybrid CBX1 recovered on tryptophan-free medium

Genotypes observed														No. observed
Class	Red side						White side							
	ADE6	LEU1	TRP5*	CYH2	MET13	ADE5	ADE6	LEU1	TRP5*	CYH2	MET13	ADE5		
1	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	6	
2	$\frac{-}{+}$	$\frac{-}{+}$	$\frac{++}{1+}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	
3	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{++}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{++}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	2	
4	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{++}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	
5	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{12}{++}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{+2}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	
6	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{12}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	
7	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{++}{+2}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	
8	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{++}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	2	
9	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{++}{+2}$	$\frac{-}{+}$	$\frac{+}{+}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	
10	$\frac{-}{+}$	$\frac{-}{+}$	$\frac{++}{+2}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{-}{+}$	$\frac{+}{-}$	$\frac{+}{-}$	$\frac{1+}{++}$	$\frac{-}{-}$	$\frac{+}{+}$	$\frac{-}{-}$	1	

* 1+ = *trp5-d* and +2 = *trp5-c*.

FIG. 1. Coincident gene conversion and reciprocal recombination on the prereplicative model of mitotic recombination (4–6, 11). (A) Recombination is initiated before replication of the homologous chromosomes because of the presence of a single-strand nick or gap in the DNA of one of the homologs. (B) Coordinated DNA synthesis (dashed line), strand displacement, and strand uptake result in asymmetric transfer of single-stranded DNA from the donor (black) to the recipient (white) homolog and transient D-loop formation. (C) The region of asymmetric heteroduplex DNA in the recipient duplex is extended by continued DNA synthesis, strand displacement, and uptake into the recipient duplex. (D) Asymmetric strand transfer terminates as the Meselson–Radding isomerization and gap closure result in the formation of a Holliday structure in which the DNA double helices of the recombining homologs are joined by crossing single DNA chains. The length of the region of asymmetric heteroduplex DNA and the site of formation of the Holliday junction are simultaneously determined. (E) Chromosomal DNA replication resolves the Holliday structure and two pairs of sister chromatids (1,2 and 3,4) are formed. Genetic markers in the region of asymmetric heteroduplex DNA exhibit coincident gene conversion. Three chromatids (nos. 1, 3, and 4) contain only the donor (black) DNA sequence of this region; one chromatid (no. 2) contains the recipient (white) DNA sequence of this region. Genetic markers bracketing the region of coincident gene conversion and the site of formation of the Holliday junction are reciprocally recombined—i.e., chromatids 1 and 4. Chromatids 2 and 3 retain the parental configuration of flanking sequences.

evidence of Holliday structure formation in the immediate vicinity of selected conversional events (i.e., Trp^+ prototrophs) and perturbation of the usual frequency distribution of conversional and reciprocal recombination events along the length of a chromosome.

In meiosis, *spoil-1/spoil-1* cells exhibit a Rec^- phenotype associated with high levels of chromosomal nondisjunction (1–3) and the *SPO11* gene product is apparently required for stable associations of nonsister chromatids (2). In meiotic yeast cells, recombination occurs postreplicatively and asymmetric heteroduplex DNA is apparently limited to short regions that are on average <1,000 base pairs long, when events involving single-copy genes are examined (15). Failure to isomerize and form Holliday structures, which are thought to be the molecular basis of meiotic chiasma formation (16), may result in disengagement of interacting chromatids in *spoil-1/spoil-1* meiotic cells. This may occur both by eventual scission of the single DNA strand uniting the interacting chromatids and by reversal of strand transfer because of branch migration of the incomplete Holliday junction to the site of single-strand scissions in the transferred strand. Branch migration can occur with considerable rapidity over a distance of $\approx 1,000$ base pairs (17).

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