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Meiotic Mapping of Yeast Ribosomal Deoxyribonucleic Acid on Chromosome XII

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We have used meiotic mapping techniques to locate the position of the repeating ribosomal DNA (rDNA) genes of the yeast *Saccharomyces cerevisiae*. We found that the rDNA genes are located on the right arm of chromosome XII, approximately 45 map units centromere distal to the gene *gal2*. Together with mapping data from previous studies, this result suggests that the tandem array of rDNA genes contains at least two junctions with the non-rDNA of the yeast chromosome. In addition, we observed segregation patterns of the rDNA genes consistent with meiotic recombination within the rDNA gene tandem array in 3 of the 59 tetrads examined.

The yeast *Saccharomyces cerevisiae* contains about 100 to 140 ribosomal DNA (rDNA) genes per haploid genome (14). Each repeating rDNA gene codes for four species of rRNA, the 25S, 18S, 5.8S, and 5S rRNA species (1, 13, 17). Within a single haploid cell, the rDNA genes are very similar (and perhaps identical) in length and base sequence (3, 12). Recent evidence suggests that most of the rDNA genes are located in a single chromosomal cluster in a tandem arrangement (10, 11).

Several years ago, evidence from several groups suggested that most of the yeast rDNA genes were located on chromosome I (4, 5, 15). The evidence for this conclusion was that the level of rDNA as measured by saturation hybridization to rRNA was lower in strains which were monosomic ($2n - 1$) for chromosome I than in normal diploid strains.

The recent discovery of a yeast strain which contained rDNA genes having a different *eco*RI restriction pattern from that observed in most laboratory strains allowed re-investigation of the chromosomal location of the rDNA genes. Most laboratory strains contain rDNA genes which have seven sites recognized by the restriction enzyme *Eco*RI (3, 12, 13); this type of rDNA has been called "form I" rDNA. A strain containing variant rDNA genes with only six sites recognized by *Eco*RI has been recently identified (12); this is the "form II" rDNA. Since form I and form II rDNA genes have different restriction patterns, agarose gel electrophoresis of *Eco*RI-treated rDNA can be used to determine whether a given strain has form I rDNA, form II rDNA, or a mixture of form I and form II. By this technique it has been shown that, within a single haploid strain, all 100 copies of the repeating rDNA genes are usually homogeneous, being either form I or form II (12).

When a haploid strain containing form I rDNA was crossed to a haploid containing form II genes, the two forms of the rDNA usually segregated in a simple Mendelian fashion (10, 11). The observation that at meiosis the rDNA genes usually segregated 2 form I spores:2 form II spores showed that most of the rDNA genes were on a single chromosome and that meiotic recombination within the rDNA gene cluster was quite rare (10, 11). Since the form I and form II rDNA genes usually behaved as alleles, this heterogeneity could be used to map the rDNA genes.

A diploid strain which was heterozygous for the form I-form II heterogeneity was constructed (T. D. Petes and S. M. Smolik-Utlaut, manuscript in preparation). Strains which were monosomic for chromosome I were selected from the diploid. Since these monosomic strains retained both form I and form II rDNA in the same proportion as the diploid, it was concluded that the rDNA genes were not located on chromosome I. The rDNA genes were then mapped by an aneuploid mapping technique and by mitotic recombination (T. D. Petes, Proc. Natl. Acad. Sci. U.S.A., in press). Results obtained by both of these methods indicated that the rDNA genes were located on chromosome XII. In addition, the mitotic recombination data indicated that the rDNA genes were located on the right arm of chromosome XII.

In this report we confirm the location of the rDNA genes on chromosome XII by using meiotic mapping techniques. In addition, we show that the rDNA genes are located approximately 45 map units centromere distal to the

gene *gal2*. Together with earlier mapping data, these results show that the rDNA gene cluster is flanked on two sides by non-ribosomal chromosomal DNA. We also found that 3 of the 59 tetrads examined in this study showed an rDNA segregation pattern consistent with meiotic recombination of the rDNA genes. This result suggests that the suppression of meiotic recombination of rDNA genes noted in earlier studies (10, 11) is not absolute.

MATERIALS AND METHODS

Strains and nomenclature. The diploid strain TP20 which was used extensively in these studies was constructed by mating the haploid strain TP12-8D (**a** *cdc14 asp5 aro7-1 trp1 ura1 his5 his2 lys11 pet17 arg4 RDN1-2* [form II rDNA]) to the haploid XY505-18C (*α his5-2 lys1-1 ura4-1 gal1 gal2 RDN1-1* [form I rDNA]). The strain XY505-18C was provided by V. MacKay (Rutgers University, New Brunswick, N.J.).

Throughout this paper, dominant alleles are indicated by capitalized 3-letter abbreviations. In most cases, the dominant allele is the wild type. Recessive alleles are indicated by lowercase-letter abbreviations. When the genetic locus is discussed without reference to a particular allele, the lowercase-letter abbreviation is used.

Genetic analysis. The procedures involved in strain construction, sporulation of diploid strains, and tetrad dissection were standard (7).

Isolation and restriction analysis of yeast rDNA. Yeast rDNA was isolated using CsCl density gradients which contained the fluorescent dye Hoechst 33258 (American Hoechst Co.) as described previously (D. H. Williamson, personal communication; 10, 11). After the gradient centrifugation, the dye is removed from the DNA by extraction with isopropanol saturated with 2 M CsCl (Petes, Proc. Natl. Acad. Sci. U.S.A., in press) and treated with the *Eco*RI restriction enzyme (Miles Research Products). Conditions used for the *Eco*RI digestion and gel electrophoresis were described previously (12).

RESULTS

Meiotic mapping of yeast rDNA genes. Aneuploid mapping techniques and mitotic recombination analysis showed that most of the yeast rDNA genes were on the right arm of chromosome XII (Petes, Proc. Natl. Acad. Sci. U.S.A., in press). In view of conflicting data indicating that the yeast rDNA genes were on chromosome I (for example, reference 4), we decided to map the rDNA genes by meiotic analysis. As will be described below, this analysis not only confirmed the previous study indicating that the rDNA genes were on chromosome XII, but allowed mapping of the rDNA genes relative to other genetic markers on this chromosome.

Several of the genetic markers previously mapped on chromosome XII are shown in Fig. 1a. To examine meiotic linkage of the rDNA genes to these markers, we constructed a diploid strain TP20 which was heterozygous for several genes previously mapped to the right arm of chromosome XII. The diploid TP20 was constructed by crossing strain TP12-8D (*asp5 GAL2 URA4* form II rDNA) to strain XY505-18C (*ASP5 gal2 ura4* form I rDNA).

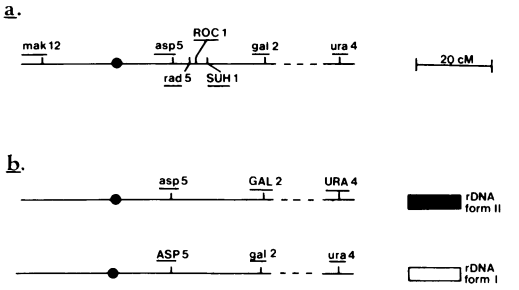

FIG. 1. *Genetic maps of chromosome XII. (a) Location of known genetic markers on chromosome XII (mapped by Mortimer and Hawthorne [8]). The gene mak12 has been mapped by R. Wickner (personal communication). (b) Parental configuration of chromosome XII markers in the diploid TP20. The asp5, GAL2, URA4, and form II rDNA genes were provided by the parental strain TP12-8D. The ASP5, gal2, ura4, and form I rDNA genes were provided by the parental strain XY505-18C. The dashed lines on the figures indicate that ura4 is unlinked to gal2 by meiotic studies but shows mitotic linkage to gal2 (8).*

The TP20 strain was sporulated, and 59 tetrads were dissected. Each spore from the tetrads was grown into a culture and checked for its genotype at the *asp5, gal2,* and *ura4* loci by plating on plates with the appropriate medium. From each culture, rDNA was isolated and treated with the *Eco*RI restriction enzyme. Since form I rDNA genes have a different *Eco*RI restriction pattern from form II rDNA genes, it was easy to determine by agarose gel electrophoresis whether a spore had form I rDNA, form II rDNA, or a mixture of form I and form II. The sensitivity of the gel system is such that about 10% contamination of form I rDNA with form II rDNA or vice versa can be detected (10). Consequently, our conclusions from the gel data are valid for at least 90% (and possibly all) of the yeast rDNA genes.

In 56 of 59 tetrads, the rDNA genes segregated two spores with form I rDNA:two spores with form II rDNA. This result confirms earlier experiments (10, 11) which indicated that the repeating rDNA genes usually behaved as a simple Mendelian unit. The 3 of 59 tetrads which did not show simple 2:2 segregation for the rDNA genes may be the result of meiotic recombination within the tandem array of repeating rDNA genes and will be discussed in more detail below.

By examining the meiotic segregation patterns of two heterozygous markers, one can determine whether the markers are genetically linked and approximately how many map units separate the two markers if they are linked (7). In yeasts, as in other systems, meiotic linkage is detected by a statistically significant excess of the parental configuration of genetic markers over the nonparental configuration after meiosis. For a diploid which is heterozygous for two genes **a** (the two alleles being **a** and **A**) and **b** (the two being **b** and **B**), three types of tetrads are usually observed. If the original parental configuration is **AB** × **ab**, **AB**, **AB**, **ab**, **ab** is parental ditype (PD), **Ab**, **Ab**, **aB**, **aB** is nonparental ditype (NPD) and **AB**, **Ab**, **aB**, **ab** is tetratype (T) (7). A statistically significant excess of PD tetrads over NPD tetrads indicates meiotic linkage. The distance between two linked markers can be calculated using the formula (9), distance in centimorgans = (T + 6NPD)/[2 (PD + NPD + T)].

The parental configuration for markers in the diploid TP20 is shown in Fig. 1b. The number of PD, NPD, and T tetrads derived from sporulating TP20 is shown for every pairwise combination in Table 1. The number of tetrads does not add up to 59 for all pairwise combinations because several of the tetrads showed departures from 2:2 segregation, presumably as the result of meiotic gene conversion (for the *gal2*, *ura4*, and *asp5* markers) or recombination (for the rDNA genes). When statistical tests were done on the data in Table 1, linkage was observed for the rDNA genes and *gal2*, the rDNA genes and *asp5*, and, as expected from earlier studies (6), linkage of *asp5* and *gal2*. The meiotic linkage between the rDNA genes and other genetic markers on chromosome XII confirms the previous mitotic recombination mapping data (Petes, Proc. Natl. Acad. Sci. U.S.A., in press).

TABLE 1. *Meiotic linkage of rDNA genes to previously mapped chromosome XII genes*[a]

Gene pair	Segregation ratio	Linkage distance (cmorgans)
ura4-rDNA	11PD:6NPD:39T	NS
gal2-rDNA	16PD:2NPD:37T	45
asp5-rDNA	16PD:4NPD:35T	54
gal2-ura4	12PD:8NPD:38T	NS
asp5-gal2	19PD:2NPD:36T	42
asp5-ura4	6PD:7NPD:45T	NS

[a] Spores from the diploid TP20 were analyzed for auxotrophic markers by plating on media lacking single nutritional requirements. rDNA was isolated from cultures grown from each spores, treated with the *Eco*RI restriction enzyme, and analyzed by agarose gel electrophoresis. Each tetrad was classified either PD (parental ditype), NPD (nonparental ditype), or T (tetratype) by examining each pair of markers separately. The definitions of PD, NPD, T are given in the text. As explained in the text, if the number of PD tetrads exceeds the number of NPD tetrads by a statistically significant amount, the markers are considered linked. NS, Not significantly linked (i.e., the difference between PD and NPD was not significant at the 95% confidence level). The map distance between linked markers was calculated using the formula derived by Perkins (9).

The calculated map distances for the three detected linkages are also given in Table 1. It should be stressed that the map distances indicated in the table are estimates which are based on the assumption that two cross-overs or less occur between the scored markers (16). Two lines of evidence suggest that the correct order of markers is: centromere-*asp5*-*gal2*-rDNA genes-*ura4*. The first is that if the rDNA genes were centromere proximal to *gal2* and 45 cmorgans from *gal2*, they should be quite closely linked to *asp5* since *gal2* and *asp5* are 42 cmorgans apart. Since the *asp5* gene is 54 cmorgans from the rDNA, the order of the genes is more likely to be: *asp5*-*gal2*-rDNA. The distance between *asp5* and the rDNA genes does not equal the sum of the *asp5*-*gal2* interval plus the *gal2*-rDNA interval. The non-additivity was presumably found because the equation used to calculated map distances underestimates long intervals (16).

A second argument which indicates that the correct map order is *asp5*-*gal2*-rDNA is based on centromere linkage data. The distance between the centromere and a heterozygous genetic marker can be determined by examining the segregation patterns of the heterozygous gene of interest with respect to a second heterozygous gene known to be closely centromere linked (7). Centromere linkage is detected by a ratio of first-division segregation tetrads (PD and NPD tetrads) to second-division segregation tetrads (T tetrads) which is significantly greater than 0.5 (7). Since the TP20 diploid was heterozygous for the tightly centromere-linked gene *trp1*, the linkage to the chromosome XII centromere of the genes *asp5*, *gal2*, *ura4* and the rDNA genes could be examined. The only significant centromere linkage was between *asp5* and the centromere (Table 2). The distance between the centromere and a gene can be determined by dividing by two the percentage of tetrads showing second-division segregation with a tightly centromere-linked marker (16). Since the percentage of second-division segregation with *asp5* was 40%, the calculated distance between *asp5* and the centromere is 20 cmorgans, compared with 13 cmorgans deter-

mined in previous experiments (6). These data support the conclusion that the rDNA genes are centromere distal to *gal2* rather than centromere proximal, since some centromere linkage of the rDNA genes would have been expected for the centromere-proximal configuration. The results also confirm the conclusion (10, 11) that the rDNA genes are not centromere linked.

The complete linkage map indicating the position of the rDNA genes is shown in Fig. 2. The distance between *asp5* and the centromere was calculated, as described above, by examining the segregation of *asp5* with respect to the centromere-linked gene *trp1*. The genes *asp5* and *gal2* were about 42 cmorgans apart, somewhat greater than the reported value of 25 cmorgans (6). The *gal2* gene is placed distal to *asp5* on the basis of previous data (6). The data given in Table 2, showing that *gal2* is not linked to the centromere, also indicate that *gal2* is centromere distal to *asp5*. The rDNA genes are located 45 cmorgans from *gal2* and are centromere distal from *gal2* as discussed previously. We propose that this genetic locus be called "*RDN1*." It should be stressed that, although most, if not all, of the rDNA genes are at this locus, the sensitivity of the gel technique is not good enough to exclude the possibility that a small fraction (less than 10%) of the rDNA genes are located elsewhere. The designation *RDN1* allows the possibility of additional chromosomal loci for a small number of rDNA genes. We further propose that the form I rDNA be designated *RDN1-1* and form II rDNA be designated as *RDN1-2*.

TABLE 2. *Tests for centromere linkage of asp5, gal2, rDNA, and ura4*[a]

Gene pair	Segregation ratio	Distance of gene from centromere (cmorgans)
trp1-asp5	32FDS:21SDS	20
trp1-gal2	18FDS:38SDS	NS
trp1-rDNA	16FDS:34SDS	NS
trp1-ura4	14FDS:44SDS	NS

[a] Centromere linkage in yeast for a given heterozygous gene can be detected by examining its meiotic segregation patterns with respect to a previously mapped centromere-linked gene (7). In TP20, the gene *trp1*, which is known to be tightly centromere linked (7), was heterozygous. The segregation patterns of the other genes in the cross with respect to *trp1* are given. Centromere linkage is indicated if the number of first-division segregation tetrads (FDS tetrads = PD + NPD tetrads) is statistically significantly more than half the number of second-division segregation tetrads (SDS tetrads = T tetrads) (7). Of the four loci examined in this study, only *asp5* was centromere linked. The distance between *asp5* and the centromere was calculated by dividing the percent SDS tetrads by 2 (16). NS, Not significant.

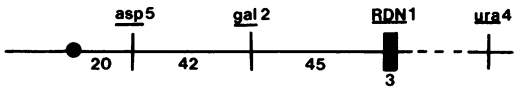

FIG. 2. *Genetic map of chromosome XII, indicating position of rDNA genes relative to other markers. Tetrads derived from TP20 were dissected. The spores were analyzed for nutritional markers and for rDNA patterns. These data were then used as described in Tables 1 and 2 to calculate gene-gene and gene-centromere linkage distances. The numbers 20, 42, 45, and 3 indicate the number of centimorgans between the centromere and asp5,asp5, and gal2,gal2, and the rDNA genes, and between the ends of the rDNA gene tandem array. The dashed line in the figure indicates that the ura4 marker is not linked meiotically to the rDNA genes and is placed on the right arm of chromosome XII as the result of other mapping results (8).*

The *RDN-1* locus is drawn as a box 3 cmorgans in length because 3 of 59 tetrads showed the patterns expected for meiotic recombination within the rDNA genes; these are discussed further below. Finally, the rDNA genes are not sufficiently close to *ura4* to show meiotic linkage (Fig. 2). The placement of *ura4* on the right arm of chromosome XII is the result of earlier mitotic recombination studies (8).

The meiotic mapping data, therefore, confirm the previous results suggesting that most of the cellular rDNA genes are on chromosome XII. In addition, the mapping shows that the rDNA genes do not go through the centomere and are not at the end of the chromosome. The rDNA gene cluster is in the middle of non-rDNA sequences on the right arm of chromosome XII and, therefore, has two junctions with non-rDNA.

Meiotic recombination of rDNA genes. In 56 of 59 tetrads derived from TP20, the rDNA genes segregated 2 form I spores from 2 form II spores. This result, which was also observed for another diploid strain +D4 (10, 11), shows that most of the rDNA genes are on a single chromosome and that meiotic recombination within the repeating genes is a rare event. The three tetrads which did not show 2:2 segregation for the rDNA heterogeneity (TP20-16, TP20-34, and TP20-61) had, in all three cases, one spore with only form I rDNA, one spore with only form II rDNA, and two spores which had both form I and II rDNA. An example of a gel analysis of one of these tetrads TP20-61 is shown in Fig. 3. The segregation pattern observed in these three tetrads is that expected if a single reciprocal meiotic exchange occurred within the tandem array of repeating rDNA genes (Fig. 4a). The segregation patterns expected for a simple

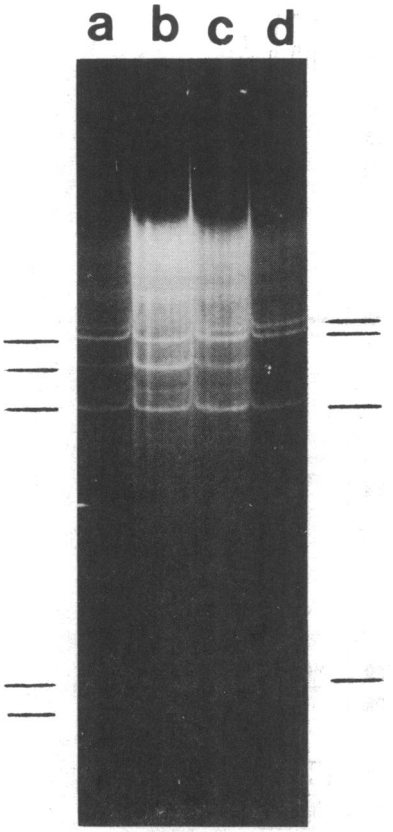

FIG. 3. *Segregation pattern of rDNA genes in a tetrad after a putative meiotic recombination event within the rDNA gene cluster. rDNA was isolated from cultures grown from the four spores of TP20-61. This rDNA was treated with the EcoRI restriction enzyme and analyzed by agarose gel electrophoresis. The restriction patterns for form I and form II rDNA are drawn on the left and right sides, respectively, of the gel. The spore TP20-61B has the form I rDNA pattern, TP20-61D has the form II rDNA pattern, and spores TP20-61A and TP20-61C each contain both form I and form II rDNA.*

mitotic recombination event (Fig. 4b and c) do not fit this pattern. If mitotic recombination of the rDNA preceded meiosis in some cells, these cells should segregate two types of spores at meiosis (Fig. 4b and c). Thus, the observation of at least three types of spores in a single tetrad (those with only form I, those with only form II, and those with both form I and form II) is most simply explained by meiotic recombination. We cannot exclude the possibility that the recombination event occurred in a mitotic cell which immediately underwent meiosis; however, this seems unlikely since none of the tetrads show the segregation pattern expected for a mitotic recombination earlier that the last mitosis before meiosis.

The genotypes (for chromosome XII markers) for the spores of the three tetrads showing putative meiotic recombination events are given in Fig. 5a through c. Also shown in Fig. 5 are the positions of the meiotic exchanges which could have produced those genotypes. In this diagram, we have attempted to indicate the minimum number of exchanges required to generate the observed genotypes. This number of exchanges is a minimum because *ura4* is too far from the rDNA gene cluster to predict the number of recombination events between *ura4* and the rDNA genes.

As mentioned previously, the meiotic recombination distance between two genetic markers can be measured using the equation: distance = [(T asci) + 6 (NPD asci)]/[2 (total)] (9). Since 3 of 59 tetrads were tetratype with respect to rDNA segregation, the recombination distance between the ends of the repeating rDNA gene cluster is about 2.5 cmorgans.

DISCUSSION

The meiotic mapping studies described here support the conclusion that most, if not all, of the yeast rDNA genes are on chromosome XII between *gal2* and *ura4*. This result indicates that there should be at least two junctions of the yeast rDNA genes with non-rDNA chromosomal sequences. We can also conclude that the yeast rDNA genes are not located near the centromere or the telomere of the yeast chromosome.

The meiotic mapping studies also confirm earlier results which indicated that the yeast rDNA usually segregated as a single Mendelian unit (10, 11). Of 59 tetrads three showed the segregation pattern expected for a single meiotic recombination event. There were, therefore, three recombination events within the rDNA genes per 59 tetrads or a frequency of 0.05 recombination events per meiotic cell.

It has been estimated that each yeast cell recombines about 70 to 100 times per meiosis (2). Since the rDNA is about 5% of the total nuclear DNA (14), the rDNA genes should recombine about 3.5 to 5 times per cell per meiosis if the frequency of recombination is proportional to physical length. Since the observed frequency of meiotic recombination is only 0.05 events per cell per meiosis, meiotic recombination appears to be suppressed by a factor of 70 to 100. This result supports earlier conclusions based on a smaller sample size (10, 11).

The data presented here show that, although meiotic recombination is suppressed by about two orders of magnitude, some meiotic recombination does occur. We cannot exclude the pos-

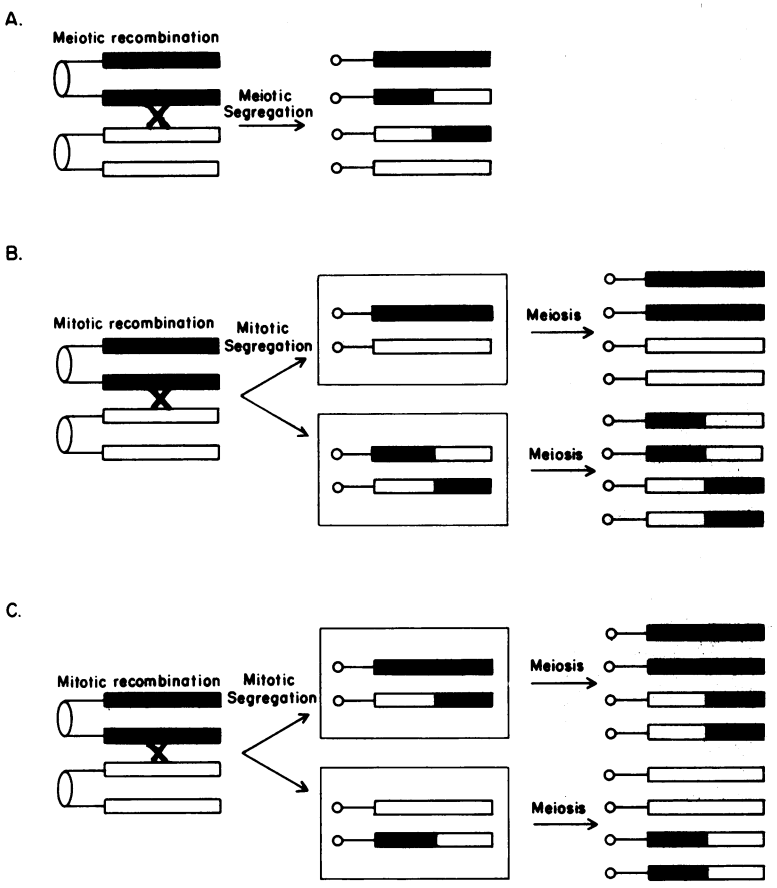

FIG. 4. *Segregation patterns expected after meiotic or mitotic recombination within the rDNA gene tandem array. For all portions of the figure, the form II rDNA gene tandem array is indicated by the black rectangle on the chromosome and the form I rDNA is indicated by the white rectangle. (A) Meiotic recombination within the rDNA genes produces one spore with only form II rDNA, one spore with only form I, and two spores with both form I and form II rDNA (B and C). Mitotic recombination occurring in cells prior to meiosis should produce only two different types of spores per tetrad (assuming that mitotic recombination does not occur in a cell which then immediately undergoes meiosis). (B) The two chromosomes which underwent the mitotic exchange segregated at mitosis into one cell and the two unrecombined chromosomes segregated into a second cell. (C) Both cells produced by the mitotic division after the mitotic exchange contained one recombined chromosome and one non-recombined chromosome.*

sibility that this recombination is occurring between non-rDNA sequences which are inserted within the rDNA gene tandem array. The lack of rDNA-non-rDNA junctions (3, 12), however, makes this possibility unlikely.

The mechanism by which meiotic recombination is suppressed is not clear. One possibility is that the rDNA genes lack a specific base sequence required to initiate recombination. A second possibility is that the rDNA genes are packaged with a set of proteins that inhibits meiotic recombination. The observation that meiotic recombination is suppressed by a factor of about 100 and there are about 100 rDNA genes in mitotic cells suggests a third possibility—that the meiotic cells contain a single copy of the rDNA genes which is amplified during spore germination. The most extreme version of this hypothesis is untenable, since a single chromosome which has a mixture of both form I and II rDNA can undergo meiosis without becoming homozygous for either form I or form II rDNA (Petes, Proc. Natl. Acad. Sci. U.S.A., in press). Furthermore, it has been demonstrated that most, if not all, of the yeast rDNA in vegetative cells is chromosomal rather than extra-chromosomal (Petes, Proc. Natl. Acad. Sci. U.S.A., in press). We cannot rule out a model in which the

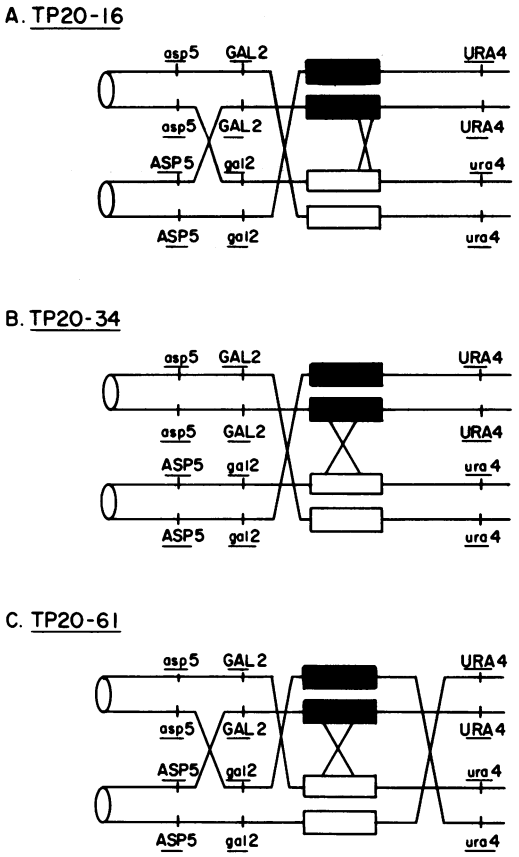

FIG. 5. *Meiotic exchanges required to produce the observed genotypes of spores from tetrads in which meiotic recombination of rDNA genes has occurred. Form II rDNA is represented by black rectangles and form I is represented by white rectangles. (A) TP20-16. Genotypes of the spores are: TP20-16a (asp5 GAL2 form I rDNA ura4), TP20-16b (ASP5 GAL2 form II > form I rDNA ura4), TP20-16c (ASP5 gal2 form II URA4), and TP20-16d (asp5 gal2 form I > form II rDNA URA4). (B) TP20-34. Genotypes of the spores are: TP20-34a (ASP5 gal2 form I and II rDNA URA4), TP20-34b (ASP5 gal2 form II rDNA URA4), TP20-34c (asp5 GAL2 form I and II rDNA ura 4), and TP20-34d (asp5 GAL2 form I rDNA ura4). (C) TP20-61. Genotypes of the spores are: TP20-61a (ASP5 GAL2 form I and II rDNA ura4), TP20-61b (ASP5 gal2 form I rDNA URA4), TP20-61c (asp5 GAL2 form I and II rDNA URA4), and TP20-61d (asp5 gal2 form II rDNA ura4).*

meiotic DNA contains a small number of rDNA genes which are later magnified by a *cis*-acting amplification mechanism. We believe, however, that suppression of meiotic recombination within a tandem array of 100 genes existing in the meiotic DNA is a simpler explanation of the data.

The rDNA segregation patterns observed in TP20 tetrads were somewhat different from the segregation patterns found in a previous study using a different diploid strain +D4 (10, 11). Among 59 tetrads from TP20, 56 showed 2:2 segregation for the rDNA genes and three showed the pattern expected for meiotic recombination. In the experiment with +D4, 12 of 14 tetrads showed 2:2 segregation for the rDNA genes and two showed the segregation patterns expected for mitotic recombination (10, 11). Calculations of the standard deviations of these measurements indicate that the differences seen in the segregation patterns of +D4 and TP20 are not statistically significant. It should be mentioned, however, that the strain +D4 has an unusually high rate of mitotic recombination (10; Petes, unpublished data), and more extensive studies with +D4 may indicate that there are quantitative differences in the frequency of mitotic (or meiotic) exchanges within the rDNA genes in different strains.

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