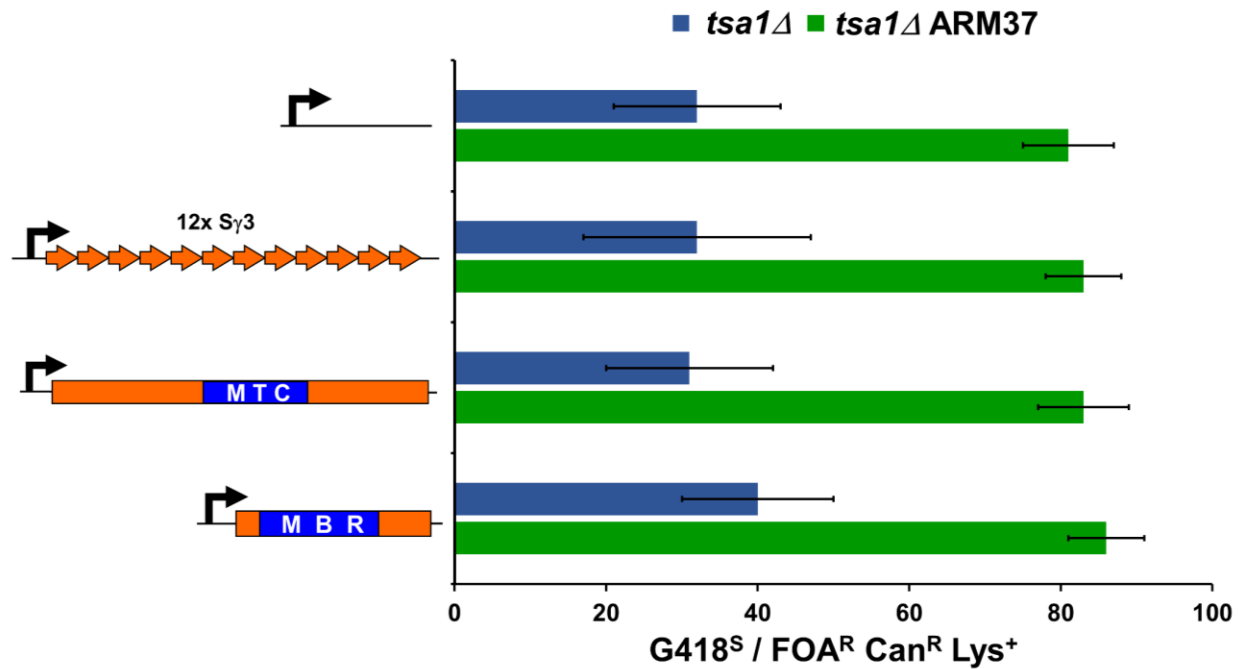


**Figure S1, Related to Figure 1. Deletion of Nearest Replication Origin has Little Effect on Recombination Rate.**

(A) Map of replication origins nearest to the assay site on Chr I with the distances of each autonomously replication sequence (ARS) (black ovals) indicated. All other symbols are the same as in Figure 1B.

(B) Recombination rate with the “No Fragile Zone” and “MBR” substrates with or without *ARS104*. These are referred to as “recombination” rate and not “ZBE” rate because the rate calculation does not subtract the G418<sup>R</sup> colonies that indicated retention of *ars104::KANMX* in order to make the two data sets comparable. Rates were determined by Luria-Delbrück fluctuation analysis and p-values by the Mann-Whitney test.

(C) Recombination rate with the 12x S $\gamma$ 3 cassetted inverted and transcribed towards the telomere by the *GAL2* promoter. While the rate in this orientation is generally higher, loss of *ARS104* has little effect. Rates determined by the method of the median with error bars representing the 95% confidence interval.



**Figure S2, Related to Figure 4. ARM37 expression Results in More Frequent Loss of the *KANMX4* Marker in *tsa1*Δ Mutants.** The total number of colonies selected for growth on medium containing 5-FOA and canavanine but lacking lysine were recorded as well as the number of these colonies that were subsequently screened for sensitivity of G418. Therefore, we can compare the percentage of 5-FOA<sup>R</sup> Can<sup>R</sup> Lys<sup>+</sup> colonies that were also G418<sup>S</sup> in *tsa1*Δ mutants with or without ARM37 expression. Error bars represent  $\pm 2$  standard error of the mean.

**Table S1, Related to STAR Methods. *S. cerevisiae* strains used in ZBE assays<sup>1,2</sup>**

<b>Strain</b>	<b>Genotype</b>
NPX416-1D	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1</i>
NPX377-4C	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p<sup>3</sup></i>
NPT126 <sup>4</sup>	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-12xS<math>\gamma</math>3</i>
NPX419-3C	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MTC</i>
NPX417-14C	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR</i>
NPX372-71B	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<math>\gamma</math>3</i>
NPX374-166B	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC</i>
NPX373-144B	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR</i>
NPX414-1A	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-RBM<sup>5</sup></i>
NPX475-XX <sup>6,7</sup>	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p, pNP171(AID, hphMX)</i>
NPX474-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<math>\gamma</math>3, pNP171(AID, hphMX)</i>
NPX473-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR, pNP171(AID, hphMX)</i>
NPX476-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC, pNP171(AID, hphMX)</i>
NPX479-XX <sup>6</sup>	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1, pNP171(AID, hphMX)</i>
NPX432-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1</i>
NPX435-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-12xS<math>\gamma</math>3</i>
NPX433-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR</i>
NPX434-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MTC</i>
NPX429-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p</i>
NPX469-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<math>\gamma</math>3</i>
NPX431-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR</i>
NPX430-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC</i>
NPX475-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p, pNP171(AID, hphMX)</i>
NPX474-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<math>\gamma</math>3, pNP171(AID, hphMX)</i>
NPX476-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC, pNP171(AID, hphMX)</i>
NPX473-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR, pNP171(AID, hphMX)</i>
NPX480-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR, pNP171(AID, hphMX)</i>
NPX381-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p</i>
NPX392-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<math>\gamma</math>3</i>
NPX380-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC</i>
NPX389-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR</i>
NPX453-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p</i>
NPX485-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<math>\gamma</math>3</i>
NPX454-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC</i>
NPX455-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR</i>
NPT148	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p, pNP171(AID, hphMX)</i>
NPT146	<i>MAT, can1::LEU2, ura3::TRP1, lys2-<math>\Delta</math>4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-</i>

GAL 1p-12xS<sub>γ</sub>3, pNP171(AID, hphMX)

NPT147 MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC, pNP171(AID, hphMX)

NPT149 MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR, pNP171(AID, hphMX)

NPX388-26B MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p

NPX394-8B MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3

NPX386-5C MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR

NPX387-5A MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC

NPX475-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p, pNP171(AID, hphMX)

NPX474-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3, pNP171(AID, hphMX)

NPX473-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR, pNP171(AID, hphMX)

NPX476-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC, pNP171(AID, hphMX)

NPX441-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p

NPX470-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3

NPX452-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR

NPX442-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC

NPX525-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p

NPX526-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3

NPX524-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC

NPX523-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR

NPT137 MATα, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p, pNP171(AID, hphMX)

NPT141 MATα, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3, pNP171(AID, hphMX)

NPT139 MATα, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC, pNP171(AID, hphMX)

NPT138 MATα, can1::LEU2, ura3::TRP1, lys2-Δ4443, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR, pNP171(AID, hphMX)

NPX529-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p

NPX530-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3

NPX528-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC

NPX527-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR

NPT144 MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p, pNP171(AID, hphMX)

NPT145 MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-12xS<sub>γ</sub>3, pNP171(AID, hphMX)

NPT143 MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MTC, pNP171(AID, hphMX)

NPT142 MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ung1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p-MBR, pNP171(AID, hphMX)

NPX421-12A MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1

NPX423-13C MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MTC

NPX422-8D MATα, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR

NPX439-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-12xS<sub>γ</sub>3

NPX438-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MTC

NPX426-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1

NPX428-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MTC

NPX427-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR

NPX438-XX MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, top1::LEU2, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MTC

NPX403-XX MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL 1p

NPX402-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC</i>
NPX404-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR</i>
NPX461-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC</i>
NPX462-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, top1::LEU2, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR</i>
NPX480-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2-Δ4443, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR, pNP171(AID, hphMX)</i>
NPX480-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR, pNP171(AID, hphMX)</i>
NPX478-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p, pNP171(AID, hphMX)</i>
NPX474-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-12xS<sub>γ</sub>3, pNP171(AID, hphMX)</i>
NPX476-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MTC, pNP171(AID, hphMX)</i>
NPX473-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-GAL1p-MBR, pNP171(AID, hphMX)</i>
NPX480-XX	<i>MAT, can1::LEU2, ura3::TRP1, lys2::ARM37, tsa1::HIS3, ars104::KANMX, bdh1::LYS2, bdh2::URA3-CAN1-MBR, pNP171(AID, hphMX)</i>
NPX220-18B	<i>MATa, can1::LEU2, ura3::TRP1, bdh2::URA3-CAN1-GAL1p, ARS104</i>
NPX205-12A	<i>MATa, can1::LEU2, ura3::TRP1, bdh2::URA3-CAN1-GAL1p, ARS104</i>
NPX246-3D	<i>MAT<math>\alpha</math>, can1::LEU2, ura3::TRP1, bdh2::URA3-CAN1-12xS<sub>γ</sub>3-GAL2p<sup>8</sup>, ARS104</i>
NPX326-5B	<i>MATa, can1::LEU2, ura3::TRP1, bdh2::URA3-CAN1-12xS<sub>γ</sub>3-GAL2p, ars104::KANMX</i>

<sup>1</sup>Strains are isogenic to W303-1A (*MATa, ade2-1, can1-100, his3-11, 15, leu2-3, 112, trp1-1, ura3-1*) with a WT *RAD5* allele. Deviations from this genotype are indicated in the table.

<sup>2</sup> Strains are listed in order of appearance in the manuscript.

<sup>3</sup> *GAL1p* = *GAL1* promoter.

<sup>4</sup> NPT strains are the result of a transformation while NPX strains are the result of a cross.

<sup>5</sup> RBM = MBR in reverse orientation.

<sup>6</sup> "XX" indicates that freshly dissect segregants were used each time from a frozen diploid stock. This is due to the strain containing a mutator allele or construct.

<sup>7</sup> Some haploids were derived from the same parent diploid.

<sup>8</sup> The *12xS<sub>γ</sub>3-GAL2p* substrate is inverted to transcribe towards the telomere and driven by the *GAL2* promoter.

**Table S2, Related to Figures 1-5. ZBE Rates ( $\times 10^{-10}$ )<sup>1</sup>**

Genotype	No Fragile Zone		12x Sy3		MTC		MBR	
	-trans	+trans	-trans	+trans	-trans	+trans	-trans	+trans
<b>Wild-type</b>	<0.48 n=20	1.8 n=20	<0.4 n=20	15.6 (4.7-26) n=20	<0.57 n=20	2.6 n=20	<0.55 n=20	1.2 n=40
<b><i>tsa1Δ</i></b>	3.5 n=19	20.3 (0-25) n=40	2.7 n=20	49 (18.9-116) n=20	2.5 n=20	13.3 (5.4-30) n=40	1.3 n=20	22.8 (9.8-36) n=60
<b>ARM37</b>	<0.42 n=20	2.9 n=40	ND <sup>2</sup>	31.4 (15-160) n=20	<0.48 n=20	2.3 n=20	<0.46 n=19	3.8 n=79
<b><i>tsa1Δ</i> ARM37</b>	ND	281 (225-357) n=60	4.2 n=20	443 (391-531) n=59	8.5 n=19	410 (310-580) n=58	ND	462 (395-558) n=60
<b><i>top1Δ</i></b>	<0.5 n=17	1.7 n=20	ND	80.3 (26-200) n=19	<0.45 n=20	2.6 n=20	<0.41 n=20	0.74 n=19
<b><i>top1Δ</i> <i>tsa1Δ</i></b>	ND	50.2 (29-91) n=39	ND	283 (239-403) n=40	6.0 n=19	56.1 (42-81) n=40	ND	80.2 (54-120) n=39
<b><i>top1Δ</i> AID</b>	ND	59.4 (25-210) n=20	ND	690 (452-939) n=20	ND	286 (196-352) n=20	ND	170 (64-312) n=20
<b><i>top1Δ</i> ARM37</b>	ND	23.2 (12-35) n=20	ND	ND	ND	5.4 (0-9.7) n=20	ND	11.2 (4.7-21) n=20
<b><i>top1Δ</i> <i>tsa1Δ</i> ARM37</b>	ND	ND	ND	ND	ND	423 (319-612) n=20	ND	373 (265-450) n=20
<b>AID</b>	<0.46 n=20	61.5 (39-91) n=73	ND	143 (93-170) n=72	ND	97.7 (72-150) n=60	<0.46 n=20	85.1 (64-100) n=74
<b>AID <i>tsa1Δ</i></b>	ND	3863 (3075-5725) n=45	ND	6070 (4031-9429) n=40	ND	7089 (4595-11320) n=60	3.8 (0-21) n=20	4147 (3000-6895) n=72
<b>AID ARM37</b>	ND	92.0 (69-110) n=79	ND	175 (140-240) n=74	ND	103 (75-157) n=60	1.0 n=20	120 (83-180) n=96
<b>AID ARM37 <i>tsa1Δ</i></b>	ND	6645 (4500-9800) n=20	ND	4304 (3047-6112) n=20	ND	4087 (3100-10000) n=20	4.9 n=20	3601 (2641-6643) n=17
<b><i>ung1Δ</i></b>	ND	0.28 n=39	ND	5.6 (2.6-10) n=20	ND	0.26 n=20	ND	0.67 n=20
<b><i>ung1Δ</i> AID</b>	ND	1.9 n=40	ND	8.6 (0-57) n=19	ND	5.8 (0-58) n=19	ND	21.6 (12-34) n=40
<b><i>ung1Δ</i> ARM37</b>	ND	2.1 n=19	ND	2.4 (0-14.9) n=20	ND	0.92 n=20	ND	0.65 n=20
<b><i>ung1Δ</i> AID ARM37</b>	ND	8.0 (0-24) n=17	ND	74.3 (47-236) n=18	ND	12.6 (0-53) n=20	ND	30.2 (9.4-47) n=40

<sup>1</sup>ZBE rates for each of the indicate substrates with or without transcription (“trans”). Rates determined by the method of the median have a 95% confidence interval in parentheses. All other rates were determined by Luria-Delbrück fluctuation analysis. The number of cultures included for each rate calculation is indicated (“n”).

<sup>2</sup>ND = Not Determined

**Table S3, Related to STAR Methods. Oligonucleotides**

<b>Oligo</b>	<b>Sequence (5'→3')</b>
NP386	GATGTACCTGTACATTGTGCAGAAGGTGAGTTTAAGCTAGGCTGTA AATGATTGTACGCG
NP387	ACTGCTTATCGATAAGATAAGTAATATCTCATTGGCTCCCTCTCCGCA GATGGCATCG
NP397	TTGCTGACCGCTTCTAGAGGCATCGCACAGTTTTAGCGAGGAAA CTTCAATAGTTTTGCCAGCGGAACGTCGTCATCCTTGTAATCC
NP398	GAGTTTTTAAGTGACATCACCCGAAAAGAAGCTAAGTCTTTCTCCTAA TTCATATTTAATTATTCTACATAGTCAGTGAGCGAGGAAGC
NP413	CTATTGAAGAGTTTTCTGCG
NP414	TGGCTATGTGATCCATGACG
NP415	GTACAATAATTAATATGAATTAGG
NP416	AAACATGAGTGGCAAGTGGC
NP428	TTTTACAATTCCAACCATGTGATGCTTGTGAGAGGGGCAGTGAAAA TCTATGTACCCACCAGCGGAATTCCAATTGC
NP429	TACTTCATTTACCGTGATTGTTAGGCGTCAATAGAATCTTAACGTT GGATTCCTTGTGGGACATATCATACGTAATGC
NP459	ATGGTCGCTCAAGTTCAAAGCAAGCTCCAACCTTTAAGAAA CGTCGTCGACGGTCATGTACGTGAGCGTATTTCC
NP460	TTATTTGTTGGCAGCTTCGAAGTATTCCTTGGAGTCTTCAACGGTTG GCTTGATGGTAGCTGTTATTTCTGGCACTTCTTGG
NP474	GATACACCATCAAATGGCTGTTTAGCTTGCCTTGTC
NP475	GATACACCAATAGGTGGGATGGCGGCGTTAGTATCG
NP493	CCCCATAAGAGGAATACTAACTCTTACCGTCGGCCTCTCTGTCCT GGATCAACAACCTGCCTGTTCCCTAGCATGTACG
NP494	GGTGCATGAGAAGATTGCCAACAAATTCAGTTATGACGGTGGCTCGA AAGAGAAAGCAAATCCATCTACCACCAGAACGG