

# GENETICS

Supporting Information

<http://www.genetics.org/cgi/content/full/genetics.109.106674/DC1>

**In *Saccharomyces cerevisiae*, yKu and Subtelomeric Core X Sequences Repress Homologous Recombination Near Telomeres as Part of the Same Pathway**

**Marcus E. Marvin, Craig D. Griffin, David E. Eyre, David B. H. Barton  
and Edward J. Louis**

Copyright © 2009 by the Genetics Society of America  
DOI: 10.1534/genetics.109.106674

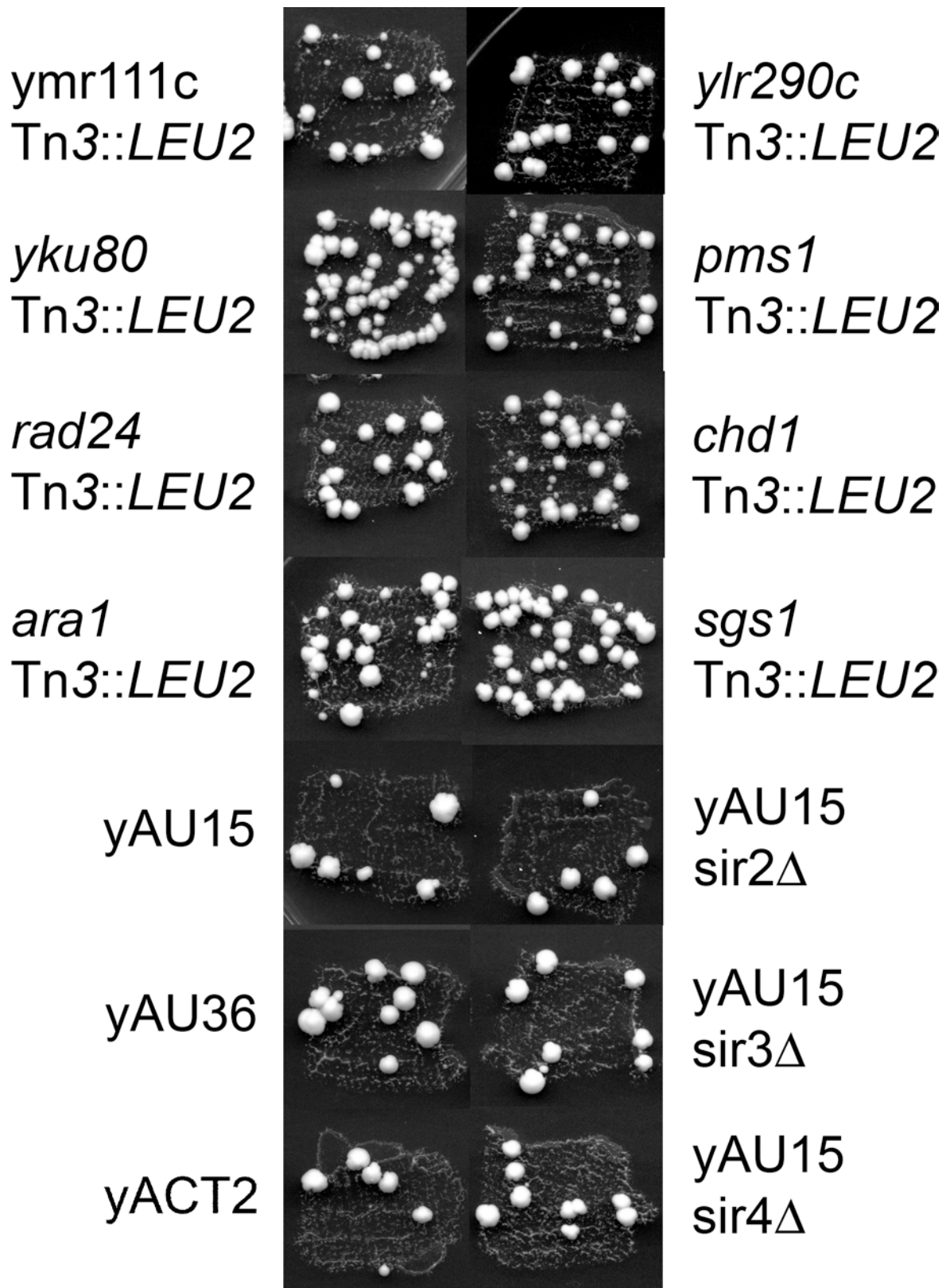


FIGURE S1.— Examples of some of the strains carrying transposon tagged alleles that confer elevated recombination in our genetic screen using strain *yAU15sir3*. It should be noted that strains carrying the *chd1*Tn3::*LEU2* allele only displayed elevated recombination when *SIR3* was also deleted in the genome, whereas the additional alleles shown above, displayed elevated recombination in all *yAU15* derived strains.

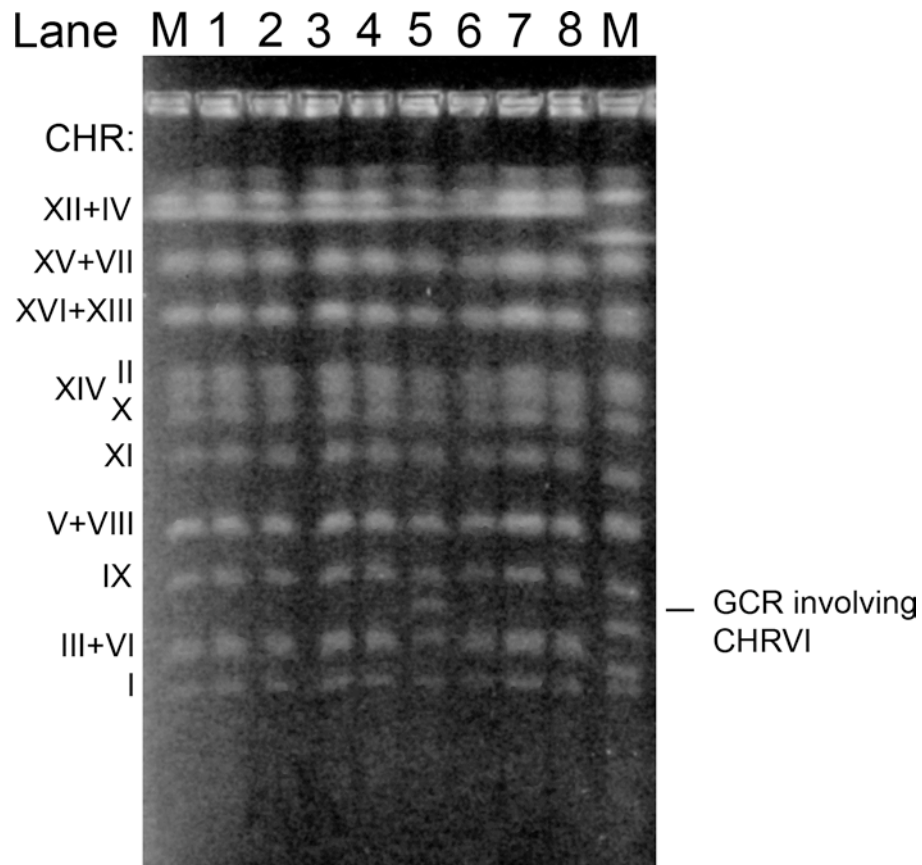


FIGURE S2.—Example of a strain from our genetic screen for hyper-recombination where a translocation involving chromosome VI had occurred. Markers (Lanes M) are the parental strain Y55 on the left hand side and S288C on the right hand side. A gross chromosomal rearrangement (GCR) involving chromosome VI can be observed in lane 5. Y55 chromosome positions on the gel are shown down the left hand side.

**FILE S1**

File S1 is available for download as an Excel file at <http://www.genetics.org/cgi/content/full/genetics.109.106674/DC1>.

**FILE S2**

**GO Annotation Correlation:** Scripts to correlate screened genes with gene ontology (GO) categories were written in Python 2.4 plus the *scipy* module. The GO term hierarchy structure was derived from the file [http://www.geneontology.org/ontology/gene\\_ontology\\_edit.obo](http://www.geneontology.org/ontology/gene_ontology_edit.obo) (revision 5.179, dated Feb 22 2007), which for each GO code gives names, definitions, top-level categories ('biological\_process', 'molecular\_function' or 'cellular\_component') and parent and child GO codes. This tree-like structure was parsed into a Python dictionary (mapping) data structure. *S. cerevisiae* gene information, including GO terms (from the *Ontology\_term* attribute) for each gene, was derived from the GFF3 formatted feature file on SGD: [ftp://genome-ftp.stanford.edu/pub/yeast/chromosomal\\_feature/saccharomyces\\_cerevisiae.gff](ftp://genome-ftp.stanford.edu/pub/yeast/chromosomal_feature/saccharomyces_cerevisiae.gff) (version dated Feb 20 2007). Each GO term in the Python data structure was further decorated with the names of all *S. cerevisiae* genes annotated with that GO term. Those GO terms not used in *S. cerevisiae* annotation were subsequently erased from the mapping. The following terms were defined: the total number of *S. cerevisiae* genes considered (N) and the number of genes highlighted in the screen (n). For each screen the following calculations were performed for each GO term in the data mapping: the total number of *S. cerevisiae* genes assigned to that GO term (D), and that measure as a proportion of the total (D/N); the total number of those genes found in the screen (k), and that measure as a proportion of the total number of genes in the screen (k/n); the ratio k/D; and the probability (p-value) of randomly achieving k hits from a sample size of n when there are a total of D possible hits in the full population size N. *Scipy*'s 'exp' and 'special.gammaln' functions were used to efficiently calculate the hyper-geometric distribution for the determination of this p-value. The same calculations were performed a second time but counting all child GO terms (and associated genes) as part of each parent GO term. The data were exported to Microsoft Excel where GO terms could be ranked by p-value (by either set of calculations), thereby highlighting which GO terms were statistically over-represented within the subpopulation of screened genes.

**FILE S3**

File S3 is available for download as an Excel file at <http://www.genetics.org/cgi/content/full/genetics.109.106674/DC1>

**TABLE S1****Strains and genetic crosses used in this study**

<sup>1</sup> Haploids (parental)	<sup>2</sup> Genotype
ND9	<i>MATa; ura3-52; ade2-101; leu2Δ; TRP1; his4-r; can1-R; CYH2-s; lys2-801</i>
ND44	<i>MATα; ura3-52; ade2-101; leu2Δ; trp1-h; HIS4; CAN1-s; cyh2-R; lys2-r2</i>
Diploids (crosses)	
PCF254 × PCF257	XVR Gap 1<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B)
PCF400 × PCF477	XVR Gap 3<(KIURA3::leu2-C) XVR Gap 3<(KIURA3::leu2-B)
PCF406 × PCF480	XVR Gap 4<(KIURA3::leu2-C) XVR Gap 4<(KIURA3::leu2-B)
PCF491 × PCF490	XVR Gap 6<(KIURA3::leu2-C) XVR Gap 6<(KIURA3::leu2-B)
PCF414 × PCF495	XVR Gap 7<(KIURA3::leu2-C) XVR Gap 7<(KIURA3::leu2-B)
PCF417 × PCF499	XVR Gap 8<(KIURA3::leu2-C) XVR Gap 8<(KIURA3::leu2-B)
PCF263 × PCF269	XVR Gap 9<(KIURA3::leu2-C) XVR Gap 9<(KIURA3::leu2-B)
PCF876 × PCF509	XVR Site 11<(KIURA3::leu2-C) XVR Site 11<(KIURA3::leu2-B)
PCF265 × PCF266	XVR Site 12<(KIURA3::leu2-C) XVR Site 12<(KIURA3::leu2-B)
PCF1126 × PCF1120	XVR Site 11<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B)
PCF1146 × PCF1129	XVR Site 12<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B)
PCF432 × PCF507	XVIL Gap 1<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B)
PCF984 × PCF1022	XVIL Site 11<(KIURA3::leu2-C) XVR Site 11<(KIURA3::leu2-B)
PCF327 × PCF334	XVIL Site 12<(KIURA3::leu2-C) XVIL Site 12<(KIURA3::leu2-B)
PCF437 × PCF504	XIIIR Gap 1<(KIURA3::leu2-C) XIIIR Gap 1<(KIURA3::leu2-B)
PCF524 × PCF1084	XIIIR Site 12<(KIURA3::leu2-C) XIIIR Site 12<(KIURA3::leu2-B)
PCF955 × PCF1013	XIIIR Site 12<(KIURA3::leu2-C) XIIIR Site 12<(KIURA3::leu2-B)
PCF941 × PCF1041	XIIIR Site 12<(KIURA3::leu2-C) XIIIR Site 12<(KIURA3::leu2-B)
PCF367 × PCF359	VIIR Site 12<(KIURA3::leu2-C) VIIR Site 12<(KIURA3::leu2-B)
PCF226 × PCF220	XIIIL Site 12<(KIURA3::leu2-C) XIIIL Site 12<(KIURA3::leu2-B)
PCF382 × PCF373	XVIR Site 12<(KIURA3::leu2-C) XVIR Site 12<(KIURA3::leu2-B)

PCF950 × PCF1009	VR Site 11<(KIURA3::leu2-C) VR Site 11<(KIURA3::leu2-B)
PCF1081 × PCF1082	VR Site 12<(KIURA3::leu2-C) VR Site 12<(KIURA3::leu2-B)
PCF1136 × PCF1111	XVR Gap 1<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1101 × PCF1117	XVR Site 12<(KIURA3::leu2-C) XVR Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1105 × PCF1138	XVIL Gap 1<(KIURA3::leu2-C) XVIL Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1114 × PCF1103	XVIL Site 12<(KIURA3::leu2-C) XVIL Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1105 × PCF1111	XVIL Gap 1<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1136 × PCF1138	XVR Gap 1<(KIURA3::leu2-C) XVIL Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1105 × PCF1117	XVIL Gap 1<(KIURA3::leu2-C) XVR Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1101 × PCF1138	XVR Site 12<(KIURA3::leu2-C) XVIL Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1114 × PCF1111	XVIL Site 12<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1136 × PCF1103	XVR Gap 1<(KIURA3::leu2-C) XVIL Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1114 × PCF1117	XVIL Gap 1<(KIURA3::leu2-C) XVR Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1101 × PCF1103	XVR Site 12<(KIURA3::leu2-C) XVIL Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1101 × PCF1111	XVR Site 12<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1136 × PCF1117	XVR Gap 1<(KIURA3::leu2-C) XVR Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1114 × PCF1138	XVIL Gap 1<(KIURA3::leu2-C) XVIL Gap 1<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1105 × PCF1103	XVIL Gap 1<(KIURA3::leu2-C) XVIL Site 12<(KIURA3::leu2-B) yku70Δ::NatMX4/yku70Δ::NatMX4
PCF1146 × PCF1129	XVR Site 12<(KIURA3::leu2-C; core X-y'acsΔ:: KanMX4) XVR Site 12<(KIURA3::leu2-B; core X-y'acsΔ:: KanMX4)
PCF1146 × PCF1129	XVR Site 12<(KIURA3::leu2-C; core X-y'acsΔ:: KanMX4) XVR Site 12<(KIURA3::leu2-B; core X-y'acsΔ:: KanMX4) yku80Δ::NatMX4/yku80Δ::NatMX4
PCF1131 × PCF1153	XVR Site 12<(KIURA3::leu2-C; Gap 8-y'acsΔ:: KanMX4) XVR Site 12<(KIURA3::leu2-B; Gap 8-y'acsΔ:: KanMX4)
PCF1131 × PCF1153	XVR Site 12<(KIURA3::leu2-C; Gap 8-y'acsΔ:: KanMX4) XVR Site 12<(KIURA3::leu2-B; Gap 8-y'acsΔ:: KanMX4) yku80Δ::NatMX4/yku80Δ::NatMX4
PCF1135 × PCF1119	XVR Site 12<(KIURA3::leu2-C) XVR Site 12<(KIURA3::leu2-B)
PCF524 × PCF490	XIIIIR Site 12<(KIURA3::leu2-C) XVR Gap 6<(KIURA3::leu2-C)
PCF524 × PCF490	XIIIIR Site 12<(KIURA3::leu2-C) XVR Gap 6<(KIURA3::leu2-B) yku80Δ::KanMX6/yku80Δ::KanMX6
PCF491 × PCF490	XVR Gap 6<(KIURA3::leu2-C) XVR Gap 6<(KIURA3::leu2-B)
PCF491 × PCF490	XVR Gap 6<(KIURA3::leu2-C) XVR Gap 6<(KIURA3::leu2-B) yku80Δ::KanMX6/yku80Δ::KanMX6



PCF876 × PCF509	XVR Site 11<(KIURA3::leu2-C) XVR Site 11<(KIURA3::leu2-B)
PCF876 × PCF509	XVR Site 11<(KIURA3::leu2-C) XVR Site 11<(KIURA3::leu2-B) yku80Δ::KanMX6/yku80Δ::KanMX6
PCF524 × PCF507	XIIIIR Site 12<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B)
PCF524 × PCF507	XIIIIR Site 12<(KIURA3::leu2-C) XVR Gap 1<(KIURA3::leu2-B) yku80Δ::KanMX6/yku80Δ::KanMX6

---

<sup>1</sup>ND9 and ND44 are the two parental strains that were used for *leu2-C* or *leu2-B* insertions. Following successful insertion of each heteroallele the parental strains were crossed to generate the diploids for analysis of recombination rates. <sup>2</sup>All diploid genotypes are therefore ND9×ND44 and denote insert sites for the *leu2* heteroalleles (see Figure 1A).

**TABLE S2****Primers used in this study**

Primer	Sequence (5'→3')
LEU2-M1	CCTAGTTAAGAACCCAACCCAggCCTAAATGGTATTATAATCACCAGC
LEU2-M2	GCTGGTGATTATAATACCATTTAGGccTGGGTTGGGTTCTTAAGTAGG
XVR-GAP 1-F1	TCAATTAAGGTTCTTGGTAATTTCTATTTCTGAAAAACCAAAACattctgggtagaagatcg
XVR-GAP 1-R1	TATGATTTTGGTAATTTGAGTCAAATAGTCAATCAATGACTTTTTTgggaataactcaggtatcg
XVR-GAP 2-F1	AAAGAAAAATAAATTTAATGTATGTATCTATATATATATATATattctgggtagaagatcg
XVR-GAP 2-R1	TAATAATAACCCAGTTGATTATGGCTATTGTCTGTTTTAAGTGATgggaataactcaggtatcg
XVR-GAP 3-F1	GATCTTGACATAAATGGCCTATTATAGAGAGTCACGCTGAACACattctgggtagaagatcg
XVR-GAP 3-R1	AGCTCTAGCAACTTAATGCAGAGAAAGATTTGAAGCTACCGCCGcgggaataactcaggtatcg
XVR-GAP 4-F1	AATTAATGATACTTATAATAAATTTGATTGAAAATAGTTACCTATTattctgggtagaagatcg
XVR-GAP 4-R1	TTCATATATATTAAGGAGTTTAACTATATACATATGATTAGGTAgggaataactcaggtatcg
XVR-GAP 5-F1	TGCAGCAAACAAAAGGGCGAAATAATAGTTACTCCAAAGATGAAattctgggtagaagatcg
XVR-GAP 5-R1	ATGCCTCCTTCGCAAACGTGTATAGTTTTCCATTTTCTTCTGCCATgggaataactcaggtatcg
XVR-GAP 6-F1	TCTTCGCCAACTCTATAAAAAGGCTGTACTCCTATAATTAAGTATgggaataactcaggtatcg
XVR-GAP 6-R1	CACAACTCATAGAAAATAAATACTAGTATGATTCCGGAAATTTattctgggtagaagatcg
XVR-GAP 7-F1	AAATGTTCTAGCTATTCATTTACCTTCCCGAAAAGCATTCTTCGAattctgggtagaagatcg
XVR-GAP 7-R1	GCAACGAAGACAGTTCTGAGATGAATGGCAGAAAGAACTGCAGCTgggaataactcaggtatcg
XVR-GAP 8-F1	TTATCCAAACCGTTGGAGCTTTCCCTTTATTTCTACATAGGTTTCattctgggtagaagatcg
XVR-GAP 8-R1	AAACAGATCGCCATAAAAAGGAGAAGCTCCGTAGGAGACCGTTTTTCgggaataactcaggtatcg
XVR-GAP 9-F1	TTTTTTCAACTACTGTTACGGTCAAGAACTAATACTGAGGATAAattctgggtagaagatcg
XVR-GAP 9-R1	TTCAAGACTTTAAATCACTTGTACCAAAATCATTGTTGCGAAAACgggaataactcaggtatcg
XVR-Site11-F1	ACGTAGATGAGCTATCGATTTTTTCTGCATACCAAGCAAGTTTACattctgggtagaagatcg
XVR-Site11-R1	ATAGATCACGCTTCAGCCGCTCTGTGTCGACTTTCTTTTCGCCAGgggaataactcaggtatcg
XVR-Site12-F1	GTATTTCACTGTTTTGATTTAGTGTGTTGTCACGGCAGTAGCGAattctgggtagaagatcg
XVR-Site12-R1	CTTTTTATAGATTGTCTTTTTATCCTACTCTTTCCACTTGTCTCgggaataactcaggtatcg
XVIL-Gap 1-F1	GATGTGGTAAAGACAACGAAATACTACGTAAATCATATTTGGTCTGattctgggtagaagatcg
XVIL-Gap 1-R1	ATTCCATTGCAGTGATTTGATTGGGAGTAAGCTTTTATAAACGAGgggaataactcaggtatcg
XIIIIR-Gap 1-F1	CATAGCTAAAGGACTATCTGGCTCGGCCATAAGTTTGAAATTTGTattctgggtagaagatcg
XIIIIR-Gap 1-R1	CTTCGACGATTTAGGTAGGTATAAGGGTTTCCCTATACCTGTATAgggaataactcaggtatcg
XIIIIR-Site12-F1	ATTGGAGGGTAACGGTTATGGTGCACGATGGGTTGGTGGTAGCAAattctgggtagaagatcg
XIIIIR-Site12-R1	TGTCCTTTCAACCATAACCGCTCCAACCACCATCCATCTCTCTACgggaataactcaggtatcg
YKU70Δ::KanMX4-F1	ATGATTTGTTAAGTGACTCTAAGCCTGATTTTAAAACGGGAATATTcgtacgctgcaggtcgac
YKU70Δ::KanMX4-R1	AAATATTGTATGTAACGTTATAGATATGAAGGATTTCAATCGTCTatcgtatgaattcaggtcgac
YKU80Δ::KanMX4-F1	AGAGTGCAGGACATATGCACAAATAATATATCTCACACCATAATAcgtacgctgcaggtcgac
YKU80Δ::KanMX4-R1	TAAGTGTGGTGACGAAAACATAACTCAAAGGATGTTAGACCTTTTatcgtatgaattcaggtcgac
KIURA3B/C-U2	CCTGGCAAACGACGATCTTC
KIURA3B/C-U3	CAATGGAACGACAGTACCCTC
XVR-GAP 1-A1	GCCGTATGTTATGGGTGCAAG
XVR-GAP 1-A4	GCTTGCTGACACAGAAGTGAC
XVR-GAP 2-A1	GCATCCTAATGCTGGTCCTAC
XVR-GAP 2-A4	GCTGCAGACATCTCTAATGTG
XVR-GAP 3-A1	CGTGATTTAGCTATCGGAACC

XVR-GAP 3-A4	CTAATACGGTAGAGTGTGCGAC
XVR-GAP 4-A1	CACTTCCTTGCCTAAAAGCTGG
XVR-GAP 4-A4	CCAGGATACATCATCGTCATC
XVR-GAP 5-A1	CGTTCCCTCTAGACATCTACAG
XVR-GAP 5-A4	ACGGTTGGTGGTTAACACCTC
XVR-GAP 6-A1	CATACCCTTGGTTGGTACGAC
XVR-GAP 6-A4	CCGTTTCGCAGTGTAAACTATG
XVR-GAP 7-A1	CGAAGGTACTACACCTCACCC
XVR-GAP 7-A4	GGCTTACGAATGAACATGAGC
XVR-GAP 8-A1	TGTCTTTTAAGAGTGAAGAGCC
XVR-GAP 8-A4	GACAGTGCAGAGTGATTTCTC
XVR-GAP 9-A1	GGGAAGGCTGTTCCAATCAAG
XVR-GAP 9-A4	CGTATTGCTAAGCCTATCTCC
XVR-Site11-A1	CAGAACTGGTGCAATGGATAG
XVR-Site11-A4	AACTGAGCCTTGCATGCAAAG
XVR-Site12-A1	CTGCAGATAACACGAGGGTAC
XVR-Site12-A4	CTCTCGCTGTCATACCTTACC
XIII-L-Site12-A4	CTTFACTCTCGCTGTCACTCC
XVIR-Site12-A4	CAAGCCCTGTTGTCTCTTACC
VIIR-Site12-A4	CAAGCCCTGTTGTCTCTTACC
XVII-Gap 1-A1	GGCACGAGTTATCCATTGCTG
XVII-Gap 1-A4	GCAAATGCACCAACAACACGG
XIIIR-Gap 1-A1	GCTAAAGTCCCATTGGTTGAC
XIIIR-Gap 1-A4	AGAAGTTCTCCTCGAGGATAG
XIIIR-Site12-A1	TAGAGTTGAGGGGATAGTGCC
XIIIR-Site12-A4	AACCACCATCCATCTCTCTAC
YKU70-A1	ACAACAGGTCACTTCTGCAAG
YKU70-A4	GGGACCCACAAAGTAATTGTC
YKU80-A1	GTAGCCTTGTGGCGCAATCG
YKU80-A4	CCTGTTTGTTCCTGGAAGTGC
KanMX4-K2	TTCAGAAACAACCTCTGGCGCA
KanMX4-K3	CATCCTATGGAACTGCCTCGG

---

Each primer is named according to its target site. LEU2-M1 and LEU2-M2 were used for site directed mutagenesis of the *KIURA3-LEU2* cassette to generate the mutant heteroalleles *KIURA3-LEU2C* and *KIURA3-LEU2B*, respectively. All primers ending with F1 or R1 were used for generating cassettes by PCR, in order to carry out targeted gene disruptions or genomic integrations. The corresponding primers to determine genomic integration at the correct locus are shown immediately to the right. Primers ending with A1, were used in conjunction with U2 or K2 primers and primers ending with A4, were used in conjunction with U3 or K2 primers, respectively.

**TABLE S3**

Table S3 is available for download as a Microsoft Word file at  
<http://www.genetics.org/cgi/content/full/genetics.109.106674/DC1>.