

1 **Table S1. Strains used in this study.**

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Strain	Parent	Genotype	Reference	
SC5314		Wild type	Gillum <i>et al.</i> 1984	
CAF2-1	SC5314	$\Delta ura3::imm434/URA3$	<i>URA3</i>	Fonzi and Irwin 1993
CAI4	CAF2-1	$\Delta ura3::imm434/\Delta ura3::imm434$	<i>ura3</i>	Fonzi and Irwin 1993
CAGL1A	CAI4	$RAD52/\Delta rad52A::hisG-URA3-hisG$	<i>RAD52, URA3</i>	This work
CAGL1A.1	CAGL1A	$RAD52/\Delta rad52A::hisG$	<i>RAD52, ura3</i>	This work
CAGL1B (TCR1)*	CAI4	$RAD52/\Delta rad52B::hisG-URA3-hisG$	<i>RAD52, URA3</i>	Ciudad <i>et al.</i> 2004
CAGL1B.1 (TCR1.1)*	CAGL1B (TCR1)	$RAD52/\Delta rad52B::hisG$	<i>RAD52, ura3</i>	Ciudad <i>et al.</i> 2004
CAGL2A	CAGL17.1	$RAD52/\Delta rad52A::hisG-URA3-hisG$ $\Delta rad59::hisG/\Delta rad59::hisG$	<i>rad52, URA3, rad59</i>	This work
CAGL2A.1	CAGL2A	$RAD52/\Delta rad52A::hisG$ $\Delta rad59::hisG/\Delta rad59::hisG$	<i>RAD52, ura3, rad59</i>	This work
CAGL2B	CAGL17	$RAD52/\Delta rad52B::hisG-URA3-hisG$ $\Delta rad59::hisG/\Delta rad59::hisG$	<i>rad52, URA3, rad59</i>	This work
CAGL2B.1	CAGL2B	$RAD52/\Delta rad52B::hisG$ $\Delta rad59::hisG/\Delta rad59::hisG$	<i>RAD52, ura3, rad59</i>	This work

CAGL3A	CAGL19	<i>RAD52/Δrad52A::hisG-URA3-hisG</i> <i>Δrad51::hisG/Δrad51::hisG</i>	<i>RAD52,</i> <i>URA3,</i> <i>rad51</i>	This work
CAGL3B	CAGL19	<i>RAD52/Δrad52B::hisG-URA3-hisG</i> <i>Δrad51::hisG/Δrad51::hisG</i>	<i>RAD52,</i> <i>URA3,</i> <i>rad51</i>	This work
CAGL4A (TCR2.1)*	CAGL1B.1	<i>Δrad52::hisG /Δrad52a::hisG-URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	Ciudad <i>et al.</i> 2004
CAGL4.1A (TCR2.2)*	CAGL1B.1	<i>Δrad52::hisG /Δrad52A::hisG-URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	Ciudad <i>et al.</i> 2004
CAGL4B	CAGL1A.1	<i>Δrad52::hisG /Δrad52B::hisG-URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	This work
CAGL5A	CAGL2B.1	<i>Δrad52/Δrad52A::hisG-URA3-hisG</i> <i>Δrad59::hisG/Δrad59::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>rad59</i>	This work
CAGL5B (TYB6.1)*	CAGL2A.1	<i>Δrad52::hisG /Δrad52B::hisG-URA3-hisG</i> <i>Δrad59::hisG/Δrad59::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>rad59</i>	Bellido <i>et al.</i> 2015
CAGL6-B (EAT2)*	EAT1.1	<i>Δrad52::hisG /Δrad52B::hisG-URA3-hisG</i> <i>Δlig4::hisG/Δlig4::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>lig4</i>	Ciudad <i>et al.</i> 2004
CAGL17.1 (BNC23.1)*	BCN1.1	<i>Δrad59::hisG/Δrad59::hisG</i>	<i>RAD52,</i> <i>ura3,</i> <i>rad59</i>	Bellido <i>et al.</i> 2015
CAGL19 (JGR5A)*	JGR5	<i>Δrad51::hisG/Δrad51::hisG</i>	<i>ura3,</i> <i>rad51</i>	García-Prieto <i>et al.</i> 2010
CAGL01 (EAT1)*	CEA2.5	<i>RAD52/Δrad52A::hisG-URA3-hisG</i> <i>Δlig4::hisG/Δlig4::hisG</i>	<i>RAD52,</i> <i>URA3,</i> <i>lig4</i>	Ciudad <i>et al.</i> 2004

EAT1.1	EAT1	<i>RAD52/Δrad52A::hisG</i> <i>Δlig4::hisG/Δlig4::hisG</i>	<i>RAD52,</i> <i>ura3, lig4</i>	Ciudad <i>et</i> <i>al.</i> 2004
CAGL4A-SAT-NTC ^R	CAGL1A	<i>Δrad52::SAT /Δrad52A::hisG-</i> <i>URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	This work
CAGL4B-SAT-NTC ^R	CAGL1B	<i>Δrad52::SAT /Δrad52B::hisG-</i> <i>URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	This work
CAGL5A-SAT-NTC ^R	CAGL2A.1	<i>Δrad52::SAT/Δrad52A::hisG-</i> <i>URA3-hisG</i> <i>Δrad59::hisG/Δrad59::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>rad59</i>	This work
CAGL5B-SAT-NTC ^R	CAGL2B.1	<i>Δrad52::SAT/Δrad52B::hisG-</i> <i>URA3-hisG</i> <i>Δrad59::hisG/Δrad59::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>rad59</i>	This work
CAGL6B-SAT-NTC ^R		<i>Δrad52::SAT/Δrad52B::hisG-</i> <i>URA3-hisG</i> <i>Δlig4::hisG/Δlig4::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>lig4</i>	This work
CAGL4A-FRT	CAGL4A-SAT-NTC ^R	<i>Δrad52::FRT /Δrad52A::hisG-</i> <i>URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	This work
CAGL4B-FRT	CAGL4B-SAT-NTC ^R	<i>Δrad52::FRT /Δrad52B::hisG-</i> <i>URA3-hisG</i>	<i>rad52,</i> <i>URA3</i>	This work
CAGL5B** ₋ FRT	CAGL5A-SAT-NTC ^R	<i>Δrad52:: FRT/Δrad52A::hisG-</i> <i>URA3-hisG</i> <i>Δrad59::hisG/Δrad59::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>rad59</i>	This work
CAGL5B-FRT	CAGL5B-SAT-NTC ^R	<i>Δrad52::FRT/Δrad52B::hisG-</i> <i>URA3-hisG</i> <i>Δrad59::hisG/Δrad59::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>rad59</i>	This work
CAGL6B-FRT	CAGL6B-SAT-NTC ^R	<i>Δrad52::FRT/Δrad52A::hisG-</i> <i>URA3-hisG</i> <i>Δlig4::hisG/Δlig4::hisG</i>	<i>rad52,</i> <i>URA3,</i> <i>lig4</i>	This work
CAGL27	CAI4	<i>RAD52/RAD52</i> <i>SHE9/she9::his-URA3-hisG</i>	<i>2 x</i>	This work

			<i>RAD52</i> , <i>URA3</i>	
CAGL28	CAGL2	<i>RAD52/rad52::hisG</i> <i>SHE9/she9::his-URA3-hisG</i>	<i>RAD52</i> <i>URA3</i>	This work

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Old names are in parenthesis. Strain CAGL5B**-FRT was intended to be CAGL5A-FRT (tester A) since it was derived from CAGL2A but behaved as if carrying *RAD52B* as the test allele. It is likely that a reciprocal exchange between both *RAD52* alleles (gene conversion or crossover) occurred at some step during its generation. NAT^R: Nourseothricin-resistant

Table S2. Primers used in this study.

Primer name	Primer sequence (5' → 3')	Size/Comments
RV1	CAACCACGACCACCACAACAA	wt: 1.6 kb <i>rad52::hisG</i> : 1.3 kb
RV2	TGCGGTATACCCGAAGAAGGA	
RV3	CGTGGCATTATGGCCTCTTC	2.5 kb
URA3.2	CGAATCGGCACTACAGC	
RV1	CAACCACGACCACCACAACAA	1.3 kb
URA3det-R	GTATGGGGTTGTTGCTCAGG	
RV2	TGCGGTATACCCGAAGAAGGA	1.8 kb
URA3.1	GGTATAGAAATGCTGCTTGG	
RAD52F- <i>Apa</i> I	GCgggcccGCTAGGTGGAAAGTGTCGTG	Small letters: <i>Apa</i> I restriction site
RAD52R- <i>Xho</i> I	GCATctcgagGGAGGTGCAGGTCTAGAG	Small letter: <i>Xho</i> I restriction site
RAD52F- <i>Sac</i> II	GCcgcggGAGGAGGTTATAGGTATC	Small letter: <i>Sac</i> II restriction site
RAD52R- <i>Sac</i> I	GTCgagctcGGGACAGTTTCAATTCAG	Small letter: <i>Sac</i> I restriction site
RAD52R	CTCAATGGTTTGGGTTATAGGATG	
SAT1F-Flip	CTGGTACTGGTTCTCGGG	
RAD52-IF	GCTGCAAGAGAGTACACAGA	
RAD52-IR	CCATAGCCAACATCTTCATG	
SNP122-F	GTGTGCTGGGTAATACGCT	227 bp
SNP122-R	GGCAAGACAATACCACAGT	

SNP123-F	GAGGAGATAGTTGTTGCTGTC	268 bp
SNP123-R	GTAGGCATTGACTACTGTGC	
SNP132-F	TCTGTCGCCTGAGCTGTTTA	256 bp
SNP132-R	CCAAACGACCCAAAATACCC	
SHE1	GAACGTGAAGTGATAGAAGACGT	wt: 845 bp
SHE2	CCCATCAAACCCAGTTCCCCAT	<i>she9::hisG</i> : 1171 bp
SHE1	GAACGTGAAGTGATAGAAGACGT	1.3 kb

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11 **Table S3** (Excel File). Molecular characterization (SNPs and PFGE-Southern blots analysis) of individual parental strains and their 5FOA^R
12 segregants analyzed in this study.