Supplemental tables and figures

Strain	Genotype/Phenotype	Ref
mcm5-461 ^{*#}	mcm5-461 ura3-52 leu2-3,112 ade2	(Dziak <i>et al.</i> , 2003)
	lys2-801 MATa	
$MCM5^*$	mcm5-461::MCM5::LEU2 ura3-52 leu2-	(Dziak <i>et al.</i> , 2003)
	3,112 ade2 lys2-801 ΜΑΤα	
<i>W303</i> [^]	ade2-1 trp1-1 can1-100 leu2-3,112 his3-	
	11,15 ura3-1 MATa	
$orc2-1^{^{^{^{^{^{^{^{^{^{^{^{^{}}}}}}}}}}}$	orc2-1 ade2-1 trp1-1 can1-100 leu2-3,112	(Ehrenhofer-Murray et al., 1995)
	his3-11,15 ura3-1 MATa	
orc5-1	orc5-1 ade2-1 trp1-1 can1-100 leu2-3,112	(Ehrenhofer-Murray et al., 1995)
	his3-11,15 ura3-1 MATa	
$cdc45-1^{\uparrow}$	cdc45-1 ade2-1 trp1-1 can1-100 leu2-3,112	(Pasero et al., 1999)
	his3-11,15 ura3-1 MATa	
<i>cdc6-1</i> ^	cdc6-1 ade2-1 trp1-1 can1-100 leu2-3,112	(Liang and Stillman, 1997)
<u>^</u>	his3-11,15 ura3-1 MATa	
$cdc7-1^{\circ}$	cdc7-1 ade2-1 trp1-1 can1-100 leu2-3,112	(Pasero <i>et al.</i> , 1999)
	his3-11,15 ura3-1 MATa ts at 37°C	
$\Delta sir2$	sir2::TRP1 ade2-1 trp1-1 can1-100 leu2-	(Rusche <i>et al.</i> , 2002)
	3,112 his3-11,15 ura3-1 MATa	
<i>LPY1030</i>	ade2-1 his3-11,15 leu2-3, 112 trp1-1 ura3-1	(Jacobson and Pillus, 2004)
	can1-100 MAT _a with telomeric	
	$adh4::URA3-UAS_{GAL}-(C_{1-3}A)_n$	
<i>LPY1029</i>	LPY1030, except lacking the telomeric	(Jacobson and Pillus, 2004)
	UAS_{GAL} tethering site	
GF100	ade2-1 trp1-1 can1-100 leu2-3,112 his3-	(Fourel <i>et al.</i> , 2002b)
	11,15 ura3-1 MATa with telomeric	
DV/7/2	$adh4::URA3-UAS_{GAL}-TRP1-STAR-(C_{1-3}A)_n$	
BY4/42	$\Delta hat I his 3\Delta I leu 2\Delta 0 lys 2\Delta 0 ura 3\Delta 0 MATa$	ATCC #4040004
$\Delta hat I$	$\Delta hat I his 3\Delta I leu 2\Delta 0 lys 2\Delta 0 ura 3\Delta 0 MATa$	ATCC#4012827
$\Delta gcn5$	$\Delta gcn \Im his \Im \Delta I \ leu 2 \Delta 0 \ lys 2 \Delta 0 \ ura \Im \Delta 0 \ MAT \alpha$	A1CC#4017285
$\Delta sas2$	$\Delta sas_2 his_3 \Delta I leu 2 \Delta 0 lys_2 \Delta 0 ura_3 \Delta 0 MAT \alpha$	ATCC#4016568
Δ sas3	$\Delta sas3 his3 \Delta I leu2 \Delta 0 lys2 \Delta 0 ura3 \Delta 0 MAT \alpha$	ATCC#4013078

Table 1. S. cerevisiae strains used in this study.

* These strains are derivatives of 8534-8C (Maine *et al.*, 1984; Hennessy *et al.*, 1991)
* This strain is identical to *cdc46-1* in (Dalton and Hopwood, 1997)
^ These strains are derivatives of *W303*** These strains are derivatives of *BY4742*

	/				<i>,</i>							
Strains	ARS1-UF	RA3-tel	ARS URA	1(AC 3-tel	S ⁻)-	ARS URA	1(B1 ⁻) 3-tel)-	URA	3-ARS1-tel	URA - tel	3-ARS1(B1 ⁻)
МСМ5	56.7 ±7.0) (n=8)	45.5	±7.8	(n=5)	89.0	±7.8	(n=5)	56.1	±7.9 (n=4)	80.0	±5.1 (n=4)
W303	56.7 ±7.9	9 (n=8)	38.4	±6.7	(n=7)	96.0	±6.7	(n=5)	33.2	±6.3 (n=9)	70.0	±7.3 (n=4)
mcm5-461	80.0 ±6.1	1 (n=8)	36.5	±3.2	(n=4)	77.0	±3.2	(n=4)	57.0	±5.3 (n=3)	81.0	±6.6 (n=3)
orc2-1	77.0 ±6.2	2 (n=6)	0.2	±0.2	(n=3)	83.0	±0.2	(n=3)	91.3	±4.3 (n=4)	88.0	±5.2 (n=4)
orc5-1	83.0 ±4.0) (n=7)	0.1	±0.1	(n=4)	83.0	±0.1	(n=4)	94.9	±3.9 (n=4)	99.0	±5.4 (n=4)
cdc45-1	81.0 ±9.0) (n=6)	28.4	±5.7	(n=4)	64.0	±5.7	(n=4)	85.1	±5.1 (n=4)	52.0	±5.9 (n=5)
⊿sas2	42.7 ±9.3	3 (n=3)	13.6	±5.7	(n=3)	n/a			n/a		n/a	
cdc7-1	63.0 ±12	.0 (n=6)	32.0	±6.9	(n=3)	86.0	±6.4	(n=4)	n/a		n/a	
cdc6-1	30.3 ±7.1	1 (n=4)	67.7	±6.4	(n=4)	86.0	±6.4	(n=4)	55.3	±9.6 (n=4)	78.0	±9.5 (n=4)
⊿sir2	0.01 ±0.0)1 (n=3)	0.02	±0.01	l (n=3)	0.02	±0.01	(n=3)	0.02	±0.01(n=3)	0.01	±0.01(n=3)

Table 2. FOA^R cells in ARS1-URA3-tel, ARS1(ACS⁻)-URA3-tel, ARS1(B1⁻)-URA3-tel, URA3-ARS1-tel, URA3-ARS1(B1⁻)-tel.

Per cent FOA^R cells in the constructs indicated above each column was measured in each of the cell strains indicated on the left. The average numbers for *n* triplicate measurements (shown in parenthesis) and the standard errors are shown.

	0 50		0 - 0	4.0.051	0 = 0	
Strains	GF6		GF6(ACS)	GF6(B1)
MCM5	48.6	±8.5 (n=5)	42.0	±2.6 (n=3)	48.0	±7.9 (n=3)
W303	63.5	±9.3 (n=3)	58.0	±7.8 (n=7)	64.4	±6.5 (n=7)
mcm5-461	9.0	±1.3 (n=6)	35.7	±2.4 (n=4)	89.7	±3.3 (n=4)
orc2-1	1.9	±0.9 (n=5)	45.5	±7.6 (n=4)	89.2	±3.5 (n=4)
orc5-1	29.2	±8.7 (n=2)	50.2	±9.8 (n=6)	90.5	±3.3 (n=6)
cdc45-1	5.3	±3.1 (n=2)	28.4	±5.5 (n=4)	31.3	±6.9 (n=4)
⊿sas2	50.9	±9.9 (n=3)	4.8	±3.0 (n=3)	n/a	
cdc7-1	34.7	±8.9 (n=3)	60.0	±6.0 (n=4)	35.6	±6.0 (n=4)
cdc6-1	61.2	±7.2 (n=5)	49.5	±5.2 (n=5)	72.2	±3.9 (n=5)
∆sir2	0.03	±0.001 (n=2)	0.02	±0.001 (n=3)	0.03	±0.001(n=3)
	GF44	ţ	GF44	(ACS ⁻)	GF44	4(B1 ⁻)
МСМ	GF44 75.5	±9.5 (n=5)	GF44 40.3	± 6.5 (n=6)	GF4 4 57.9	4(B1⁻) ±5.1 (n=6)
МСМ W303	GF44 75.5 68.5	±9.5 (n=5) ±9.7 (n=3)	GF44 40.3 41.1	±6.5 (n=6) ±7.9 (n=9)	GF4 4 57.9 51.8	4(B1⁻) ±5.1 (n=6) ±6.0 (n=9)
MCM W303 mcm5-461	GF44 75.5 68.5 9.1	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6)	GF4 4 40.3 41.1 35.7	±6.5 (n=6) ±7.9 (n=9) ±6.3 (n=5)	GF4 4 57.9 51.8 75.0	4(B1⁻) ±5.1 (n=6) ±6.0 (n=9) ±5.2 (n=5)
MCM W303 mcm5-461 orc2-1	GF44 75.5 68.5 9.1 31.8	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6) ±1.1 (n=3)	GF44 40.3 41.1 35.7 47.6	±6.5 (n=6) ±7.9 (n=9) ±6.3 (n=5) ±5.6 (n=5)	GF44 57.9 51.8 75.0 61.3	4(B1') ±5.1 (n=6) ±6.0 (n=9) ±5.2 (n=5) ±7.2 (n=5)
MCM W303 mcm5-461 orc2-1 orc5-1	GF44 75.5 68.5 9.1 31.8 19.6	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6) ±1.1 (n=3) ±7.1 (n=2)	GF44 40.3 41.1 35.7 47.6 37.3	±6.5 (n=6) ±7.9 (n=9) ±6.3 (n=5) ±5.6 (n=5) ±4.2 (n=8)	GF44 57.9 51.8 75.0 61.3 63.4	4(B1') ±5.1 (n=6) ±6.0 (n=9) ±5.2 (n=5) ±7.2 (n=5) ±7.3 (n=8)
MCM W303 mcm5-461 orc2-1 orc5-1 cdc45-1	GF44 75.5 68.5 9.1 31.8 19.6 7.5	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6) ±1.1 (n=3) ±7.1 (n=2) ±4.0 (n=2)	GF4 4 40.3 41.1 35.7 47.6 37.3 56.1	±6.5 (n=6) ±7.9 (n=9) ±6.3 (n=5) ±5.6 (n=5) ±4.2 (n=8) ±6.8 (n=4)	GF4 4 57.9 51.8 75.0 61.3 63.4 62.8	2(B1) ±5.1 (n=6) ±6.0 (n=9) ±5.2 (n=5) ±7.2 (n=5) ±7.3 (n=8) ±5.7 (n=4)
MCM W303 mcm5-461 orc2-1 orc5-1 cdc45-1 ⊿sas2	GF44 75.5 68.5 9.1 31.8 19.6 7.5 53.1	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6) ±1.1 (n=3) ±7.1 (n=2) ±4.0 (n=2) ±8.9 (n=3)	GF4 4 40.3 41.1 35.7 47.6 37.3 56.1 8.5	$\begin{array}{c} \underline{(ACS')} \\ \pm 6.5 (n=6) \\ \pm 7.9 (n=9) \\ \pm 6.3 (n=5) \\ \pm 5.6 (n=5) \\ \pm 4.2 (n=8) \\ \pm 6.8 (n=4) \\ \pm 4.3 (n=3) \end{array}$	GF44 57.9 51.8 75.0 61.3 63.4 62.8 n/a	4(B1') ±5.1 (n=6) ±6.0 (n=9) ±5.2 (n=5) ±7.2 (n=5) ±7.3 (n=8) ±5.7 (n=4)
MCM W303 mcm5-461 orc2-1 orc5-1 cdc45-1 Дsas2 cdc7-1	GF44 75.5 68.5 9.1 31.8 19.6 7.5 53.1 36.6	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6) ±1.1 (n=3) ±7.1 (n=2) ±4.0 (n=2) ±8.9 (n=3) ±9.1 (n=2)	GF4 4 40.3 41.1 35.7 47.6 37.3 56.1 8.5 62.0	$\begin{array}{c} \underline{(ACS')} \\ \pm 6.5 (n=6) \\ \pm 7.9 (n=9) \\ \pm 6.3 (n=5) \\ \pm 5.6 (n=5) \\ \pm 4.2 (n=8) \\ \pm 6.8 (n=4) \\ \pm 4.3 (n=3) \\ \pm 2.6 (n=4) \end{array}$	GF44 57.9 51.8 75.0 61.3 63.4 62.8 n/a 55.9	4(B1) ±5.1 (n=6) ±6.0 (n=9) ±5.2 (n=5) ±7.2 (n=5) ±7.3 (n=8) ±5.7 (n=4) ±9.2 (n=6)
MCM W303 mcm5-461 orc2-1 orc5-1 cdc45-1 Δsas2 cdc7-1 cdc6-1	GF44 75.5 68.5 9.1 31.8 19.6 7.5 53.1 36.6 27.9	±9.5 (n=5) ±9.7 (n=3) ±3.7 (n=6) ±1.1 (n=3) ±7.1 (n=2) ±4.0 (n=2) ±8.9 (n=3) ±9.1 (n=2) ±9.4 (n=5)	GF4 4 40.3 41.1 35.7 47.6 37.3 56.1 8.5 62.0 55.1	$\begin{array}{c} \underline{(ACS')} \\ \pm 6.5 (n=6) \\ \pm 7.9 (n=9) \\ \pm 6.3 (n=5) \\ \pm 5.6 (n=5) \\ \pm 4.2 (n=8) \\ \pm 6.8 (n=4) \\ \pm 4.3 (n=3) \\ \pm 2.6 (n=4) \\ \pm 5.6 (n=4) \end{array}$	GF44 57.9 51.8 75.0 61.3 63.4 62.8 n/a 55.9 41.5	$\begin{array}{c} \underline{4(B1')} \\ \pm 5.1 \text{ (n=6)} \\ \pm 6.0 \text{ (n=9)} \\ \pm 5.2 \text{ (n=5)} \\ \pm 7.2 \text{ (n=5)} \\ \pm 7.3 \text{ (n=8)} \\ \pm 5.7 \text{ (n=4)} \\ \pm 9.2 \text{ (n=6)} \\ \pm 5.6 \text{ (n=4)} \end{array}$

Table 3. FOA^R cells in GF6, GF6(ACS⁻), GF6(B1⁻), GF44, GF44(ACS⁻) GF44(B1⁻).

Per cent FOA^R cells in the constructs indicated above each column was measured in each of the cell strains indicated on the left. The average numbers for n triplicate measurements (shown in parenthesis) and the standard errors are shown.

Table 4. FOA	cells in GF6	(ACSASTAR)	and GF6(B1 /	ASTAR).

Strains	GF6(ACS ⁻ ASTAR)	GF6(B1 ⁻ ∆STAR)
МСМ5	64.5	±8.6 (n=3)	70.2	±6.4 (n=3)
W303	52.2	±9.8 (n=3)	55.1	±8.3 (n=3)
mcm5-461	58.3	±8.3 (n=3)	76.8	±7.1 (n=3)
orc2-1	20.1	±3.0 (n=6)	30.8	±7.2 (n=6)
orc5-1	35.5	±7.4 (n=6)	41.0	±7.0 (n=6)
cdc45-1	40.1	±7.9 (n=5)	53.2	±7.2 (n=5)

Per cent FOA^R cells in the constructs indicated above each column was measured in each of the cell strains indicated on the left. The average numbers for n triplicate measurements (shown in parenthesis) and the standard errors are shown.

URA3-UAS _{GAL4} -tel	%FOA ^R Cells
GBD	22.0 ±4.9 (n=3)
GBD-ORC1	23.1 ±7.8 (n=3)
GBD-ORC2	39.3 ±9.8 (n=3)
GBD-MCM10	86.5 ±8.3 (n=3)
GBD-mcm10-1	57.2 ±6.5 (n=3)
GBD-SIR1	75.7 ±6.3 (n=3)
GBD-MCM5	5.5 ±2.0 (n=3)
GBD-mcm5-1	8.8 ±2.8 (n=3)
GBD-mcm5-461	10.6 ±3.2 (n=3)
GBD-mcm5(bob1-1)	28.8 ±13.4 (n=6)
GBD-MCM3	56.3 ±7.9 (n=3)
URA3-tel(LPY1029)	
GAL4DBD-Orc1p	23.4 ±11 (n=6)
GAL4DBD-Mcm5-461p	58.9 ±7 (n=6)
GAL4DBD-Mcm5-1p	40.9 ±14 (n=6)
GAL4DBD-Mcm5p	19.3 ±4 (n=6)
GAL4DBDp	59.0 ±10 (n=6)
URA3- UAS _{GAL4} -tel(LPY1030)	
GAL4DBD-Orc1p	58.60 ±12 (n=6)
GAL4DBD-Mcm5-461p	1.94 ±0.4 (n=6)
GAL4DBD-Mcm5-1p	0.80 ±0.1 (n=6)
GAL4DBD-Mcm5p	0.61 ±0.1 (n=6)
GAL4DBDp	36.39 ±10 (n=6)

 Table 5A. FOA^R in W303/URA-UAS_{GAL}-tel, LPY1029 and LPY1030 cells expressing

 GBD-fusion proteins.

Per cent FOA^R cells transformed with vectors expressing the recombinant proteins presented on the left. The average numbers for *n* triplicate measurements (shown in parenthesis) and the standard errors are shown. W303/ URA-UAS_{GAL}-tel contains four UAS_{GAL} sites, while LPY1030 contains one UAS_{GAL} site.

Table 5B. FOA^{R} in HAT deletion mutants harboring URA-UAS_{GAL}-tel and expressing GBD or GBD-Mcm5p.

Strains	Protein	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
∆hat1	GBD	95.1 ±3.1(n=3)
	GBD-Mcm5p	93.2 ±3.4(n=3)
∆gcn5	GBD	84.7 ±7.1(n=3)
	GBD-Mcm5p	85.6 ±5.6(n=3)
∆sas2	GBD	94.3 ±3.1(n=3)
	GBD-Mcm5p	53.7 ±7.4(n=3)
∆sas3	GBD	95.6 ±2.8(n=3)
	GBD-Mcm5p	36.9 ±7.2(n=3)
wt	GBD	75.8 ±8.7(n=3)
	GBD-Mcm5p	13.7 ±6.2(n=3)

The average %FOA^R for *n* triplicate measurements (shown in parenthesis) and the standard errors are shown.

GF100	% FOA	R	%TRP	+	TRP+/	FOA ^R
GAL4DBD-GCN5	0.31	±0.10 (n=6)	80.01	±5.1 (n=6)	0.04	±0.02 (n=6)
GAL4DBD-ORC1	8.17	±0.59 (n=6)	36.23	±3.3 (n=6)	0.01	±0.01 (n=6)
GAL4DBD-mcm5-461	0.22	±0.17 (n=6)	94.1	±8.8 (n=6)	0.02	±0.01 (n=3)
GAL4DBD-mcm5-1	0.38	±0.15 (n=6)	84.5	±18.7 (n=6)	0.04	±0.01 (n=3)
GAL4DBD-MCM5	0.71	±0.37 (n=6)	91.4	±1.2 (n=6)	0.03	±0.004 (n=4)
GAL4DBD	3.42	±0.26 (n=6)	68.5	±4.0 (n=6)	0.01	±0.01(n=6)
GF100∆rif1	% FOA	R	%TRP	+	TRP+/	FOA ^R
GF100∆rif1 GAL4DBD-GCN5	% FOA 23.02	^R ±2.02 (n=6)	%TRP 48.4	+ ±6.2(n=6)	TRP+/ 0.8	FOA ^R ±0.1 (n=6)
GF100∆rif1 GAL4DBD-GCN5 GAL4DBD-ORC1	% FOA 23.02 74.9	^R ±2.02 (n=6) ±3.18 (n=6)	%TRP 48.4 5.01	+ +6.2(n=6) +1.9 (n=6)	TRP+/ 0.8 0.4	FOA ^R ±0.1 (n=6) ±0.05 (n=6)
GF100∆rif1 GAL4DBD-GCN5 GAL4DBD-ORC1 GAL4DBD-mcm5-461	% FOA 23.02 74.9 57.6	x ^R ±2.02 (n=6) ±3.18 (n=6) ±6.41 (n=6)	%TRP 48.4 5.01 19.6	+ ±6.2(n=6) ±1.9 (n=6) ±3.1 (n=6)	TRP+/ 0.8 0.4 0.4	FOA^{R} ±0.1 (n=6) ±0.05 (n=6) ±0.1 (n=6)
GF100∆rif1 GAL4DBD-GCN5 GAL4DBD-ORC1 GAL4DBD-mcm5-461 GAL4DBD-mcm5-1	% FOA 23.02 74.9 57.6 51.4	x ^R ±2.02 (n=6) ±3.18 (n=6) ±6.41 (n=6) ±3.86 (n=6)	%TRP 48.4 5.01 19.6 29.2	+ ±6.2(n=6) ±1.9 (n=6) ±3.1 (n=6) ±1.9 (n=6)	TRP+/ 0.8 0.4 0.4 1.4	FOA ^R $\pm 0.1 (n=6)$ $\pm 0.05 (n=6)$ $\pm 0.1 (n=6)$ $\pm 0.2 (n=6)$
GF100∆rif1 GAL4DBD-GCN5 GAL4DBD-ORC1 GAL4DBD-mcm5-461 GAL4DBD-mcm5-1 GAL4DBD-MCM5	% FOA 23.02 74.9 57.6 51.4 23.7	$\frac{\pm 2.02 (n=6)}{\pm 3.18 (n=6)}$ $\pm 6.41 (n=6)$ $\pm 3.86 (n=6)$ $\pm 2.12 (n=6)$	%TRP 48.4 5.01 19.6 29.2 45.6	$\begin{array}{c} \pm \\ \pm 6.2(n=6) \\ \pm 1.9 (n=6) \\ \pm 3.1 (n=6) \\ \pm 1.9 (n=6) \\ \pm 6.2 (n=6) \end{array}$	TRP+/ 0.8 0.4 0.4 1.4 1.0	FOA ^R $\pm 0.1 (n=6)$ $\pm 0.05 (n=6)$ $\pm 0.1 (n=6)$ $\pm 0.2 (n=6)$ $\pm 0.1 (n=6)$

Table 6. FOA^R, TRP+ and FOA^R&TRP+ cells in *GF100* and *GF100* Δ *rif1*.

Cells transformed with vectors expressing the recombinant proteins presented on the left are shown. The average numbers for n triplicate measurements (shown in parenthesis) and the standard errors are shown.

Supplemental Figure # 1:

A) The sequence of *MCM5(CDC46)* is shown. The positions of the mutations in *mcm5-461* (G549) and *mcm5-1* (G1981) are shown in rectangles and bold script. B) The amino acid sequence of Mcm5p is shown. The Zn-finger domain (amino acid residues 183-235) and the conserved heptad K/R repeat (amino acid residues 651-664) are highlighted in grey. The ATPase motif (MCM box) is underlined. The positions of the amino acid substitutions in Mcm5-461p (C183Y) and Mcm5-1p (E661K) are in rectangles and bold script.

Comments to Supplemental Figure 1: The mutation in *mcm5-461* (C183Y) is identical to the previously reported *cdc46-1* mutation (Dalton and Hopwood, 1997) and locates to the Zn-finger domain. It affects the interaction of Mcm5p with Mcm7p and the assembly of the MCM hexamer (Dalton and Hopwood, 1997). By homology with the archeal Mcm protein, this mutation is also expected to interfere with the association of the MCM hexamer with DNA (Fletcher *et al.*, 2003). The E661K mutation in *mcm5-1* locates in the conserved heptad K/R repeat. It has been postulated that this repeat provides a surface for interaction with other proteins, including other Mcm proteins (Kubota *et al.*, 1997).

A)

1	ATGTCATTTG	ATAGACCGGA	AATATACAGT	GCTCCTGTTT	TACAAGGAGA
51	ATCTCCTAAC	GACGATGATA	ATACTGAAAT	CATAAAGTCC	TTTAAGAATT
101	TCATTTTGGA	GTTCAGACTT	GACTCGCAAT	TTATTTACAG	AGATCAGTTA
151	AGGAACAACA	TCCTTGTGAA	GAATTATTCT	TTAACGGTTA	ACATGGAGCA
201	TTTGATCGGA	TATAACGAAG	ACATATATAA	GAAACTATCA	GACGAACCTT
251	CAGATATCAT	TCCATTATTC	GAAACCGCGA	TCACACAAGT	GGCTAAAAGG
301	ATAAGTATTC	TAAGCAGAGC	TCAATCTGCT	AATAACAATG	ACAAAGATCC
351	AGAAAATACT	AGTATGGATA	CTGATTCTCT	CTTATTGAAC	TCTTTACCAA
401	CATTTCAATT	AATTTTAAAC	TCCAATGCAA	ATCAGATTCC	ATTGAGAGAT
451	TTGGATTCTG	AACACGTCTC	CAAGATTGTC	CGTTTATCAG	GTATTATAAT
501	ATCCACGTCA	GTTTTATCTT	CCCGTGCCAC	GTACCTTTCT	ATAATGT G CA
551	GAAATTGCAG	ACACACAACA	TCAATAACAA	TCAACAATTT	CAATTCTATC
601	ACAGGCAATA	CCGTCAGTTT	ACCACGTTCT	TGCTTATCTA	CGATTGAGAG
651	TGAATCTTCT	ATGGCAAACG	AGTCGAATAT	TGGTGATGAA	TCGACCAAGA
701	AAAATTGTGG	ACCTGATCCA	TATATTATTA	TCCATGAGTC	TTCAAAGTTT
751	ATTGATCAAC	AGTTTTTAAA	ATTACAGGAA	ATCCCAGAAC	TGGTTCCAGT
801	AGGTGAGATG	CCTAGAAACT	TAACAATGAC	TTGTGACCGA	TACCTAACAA
851	ACAAAGTTAT	TCCTGGTACG	AGAGTCACTA	TAGTAGGTAT	TTATTCCATC
901	TATAATTCTA	AAAATGGTGC	CGGATCCGGA	AGGAGCGGGG	GTGGAAATGG
951	AGGAAGTGGT	GTTGCTATTA	GAACACCTTA	TATCAAAATA	TTAGGTATTC
1001	AGTCCGATGT	AGAAACCTCC	TCTATTTGGA	ATTCAGTAAC	TATGTTTACT
1051	GAGGAGGAAG	AAGAGGAATT	TCTACAGCTA	AGTAGAAACC	CGAAGCTTTA
1101	TGAAATTTTG	ACCAACTCTA	TTGCCCCCTC	TATTTTTGGT	AATGAAGATA
1151	TAAAAAAGC	CATTGTATGT	TTATTGATGG	GTGGTTCCAA	GAAGATATTA
1201	CCCGATGGTA	TGAGGTTAAG	AGGTGATATC	AATGTACTAT	TATTAGGTGA
1251	TCCAGGTACC	GCCAAATCTC	AACTATTGAA	ATTTGTGGAG	AAAGTGTCAC
1301	CTATTGCGGT	ATATACATCT	GGTAAGGGAT	CTTCTGCAGC	TGGGTTAACT

1351	GCCAGTGTAC	AAAGAGATCC	GATGACAAGA	GAATTTTATT	TGGAAGGTGG
1401	TGCTATGGTG	CTTGCCGATG	GTGGTGTTGT	ATGCATCGAT	GAATTCGATA
1451	AAATGAGAGA	TGAAGATAGA	GTGGCCATTC	ATGAAGCTAT	GGAGCAGCAA
1501	ACAATCTCCA	TCGCAAAAGC	TGGTATCACT	ACAGTGCTAA	ATTCTAGAAC
1551	TAGTGTTTTA	GCGGCTGCTA	ATCCGATATA	CGGCCGGTAT	GATGATTTGA
1601	AGTCTCCTGG	TGACAACATT	GATTTCCAAA	CTACTATTTT	ATCCCGTTTT
1651	GATATGATTT	TTATTGTTAA	GGATGACCAT	AATGAAGAAC	GTGATATTTC
1701	AATAGCTAAC	CACGTTATTA	ATATTCATAC	AGGAAATGCT	AATGCTATGC
1751	AAAACCAACA	AGAGGAAAAT	GGCAGTGAAA	TTAGTATTGA	AAAGATGAAA
1801	CGTTACATTA	CGTATTGTAG	ATTGAAATGT	GCACCAAGAC	TTTCACCGCA
1851	GGCCGCTGAA	AAACT <i>GTCAT</i>	CGAACTTCGT	CACCATTAGG	AAGCAATTAT
1901	TAATCAACGA	ATTAGAGTCA	ACGGAAAGGT	CGTCTATTCC	AATTACCATT
1951	CGTCAATTAG	AAGCTATTAT	TAGAATAACA	G AATCATTAG	CCAAGTTAGA
2001	ATTAAGTCCT	ATTGCACAGG	AGAGACATGT	TGACGAAGCT	ATTAGATTGT
2051	TTCAAGCTTC	CACAATGGAC	GCAGCGTCTC	AGGATCCAAT	TGGCGGCTTA
2101	AATCAAGCAA	GCGGAACATC	TTTGTCAGAA	ATCCGTCGTT	TTGAACAAGA
2151	ACTAAAAAGA	AGATTACCTA	TAGGCTGGTC	TACTTCTTAT	CAAACTTTGA
2201	GGAGAGAATT	TGTAGATACA	CATAGATTTT	CTCAATTAGC	ACTGGATAAG
2251	GCCTTATATG	CCCTAGAGAA	GCATGAAACA	ATTCAATTGA	GACACCAGGG
2301	ACAGAATATT	TACAGAAGTG	GTGTATGA		

B)

1 MSFDRPEIYSAPVLQGESPNDDDNTEIIKSFKNFILEFRL 40 41 DSQFIYRDQLRNNILVKNYSLTVNMEHLIGYNEDIYKKL 79 80 SDEPSDIIPLFETAITQVAKRISILSRAQSANNNDKDPENTS121 122 M D T D S L L L N S L P T F Q L I L N S N A N Q I P L R D L D S E H V S K I V R 161 162 LSGIIISTSVLSSRATYLSIMCRNCRHTTSITINNFNSITG 201 202 N T V S L P R S C L S T I E S E S S Met A N E S N I G D E S T K K N C G P D P Y I 241 242 IIHESSKFIDQQFLKLQEIPELVPVGEMPRNLTMTCDRY 280 281 LTNKVIPGTRVTIVGIYSIYNSKNGAGSGRSGGGNGGSGV 320 321 AIRTPYIKILGIQSDVETSSIWNSVTMFTEEEEEEFLQLS 360 361 RNPKLYEILTNSIAPSIFGNEDIKKAIVCLLMGGSKKILP 400 401 DG M RLRGDINVLLLGDPGTAKSQLLKFVEKVSPIAVYTS 439 440 GKGSSAAGLTASVQRDPMTREFYLEGGAMVLADGGVV 476 477 CIDEFDK MRDEDRVAIHEA MEQQTISIAKAGITTVLNS 514 515 R T S V L A A A N P I Y G R Y D D L K S P G D N I D F Q T T I L S R F D M I F I 554 555 VKDDHNEERDISIANHVINIHTGNANAMQNQQEENGSEI 593 594 SIEK MKRYITYCRLKCAPRLSPQAAEKLSSNFVTIRKQLL 633 634 INELESTERSSIPITIRQLEAURITESLAKLELSPIAQERHV 676 677 DEAIRLFQAST M DAASQDPIGGLNQASGTSLSEIRRFEQE 716 717 LKRRLPIGWSTSYQTLRREFVDTHRFSQLALDKALYALEK 756 757 H E T I Q L R H Q G Q N I Y R S G V Stop 775

Supplemental Figure #2

- Wild type *W303* or *cdc7-1* cells harboring URA3-UAS_{GAL}-tel (shown on the top) were transformed with plasmids carrying expression cassettes for the GBD-fusion proteins shown on the left. Per cent FOA^R for each recombinant protein was acquired and average value and error are shown on the right. The ratio %FOA^R_{GAL4-fusion} /%FOA^R_{GAL4DBD} was calculated and plotted in the graph. Values below 1 indicate that the tethered proteins have anti-silencing activity and values above 1 indicate silencing activity.
- **Comments to Supplemental Figure #2.** On-going research in our lab has shown that cdc7-1 cells are very slow in exiting from repressed or de-repressed state at the telomere (slow recovery from SC-ura or SC/FOA plates, respectively). For this reason the experiments in cdc7-1 were performed after selection on either SC-ura and SC/FOA plates followed by growth in non-selective medium to maintain the expressing plasmids but allow variegation of URA3. After 40 generations the assessment of % FOA was performed. Mcm5p does not show any anti-repression activity in cdc7-1 regardless of the initial selection. Under the same experimental conditions Orc1p still acts as a repressor after initial selection was on SC-ura. We therefore believe that the lack of silencing activity in cdc7-1 is not a consequence of the slow recovery phenotype but reflects an independent deficiency of the cdc7-1 cells.

Additional note: the silencing activity of Orc1p is not evident after selection on SC/FOA because of the already high level (75%) of repression.

