SUPPLEMENTAL MATERIAL

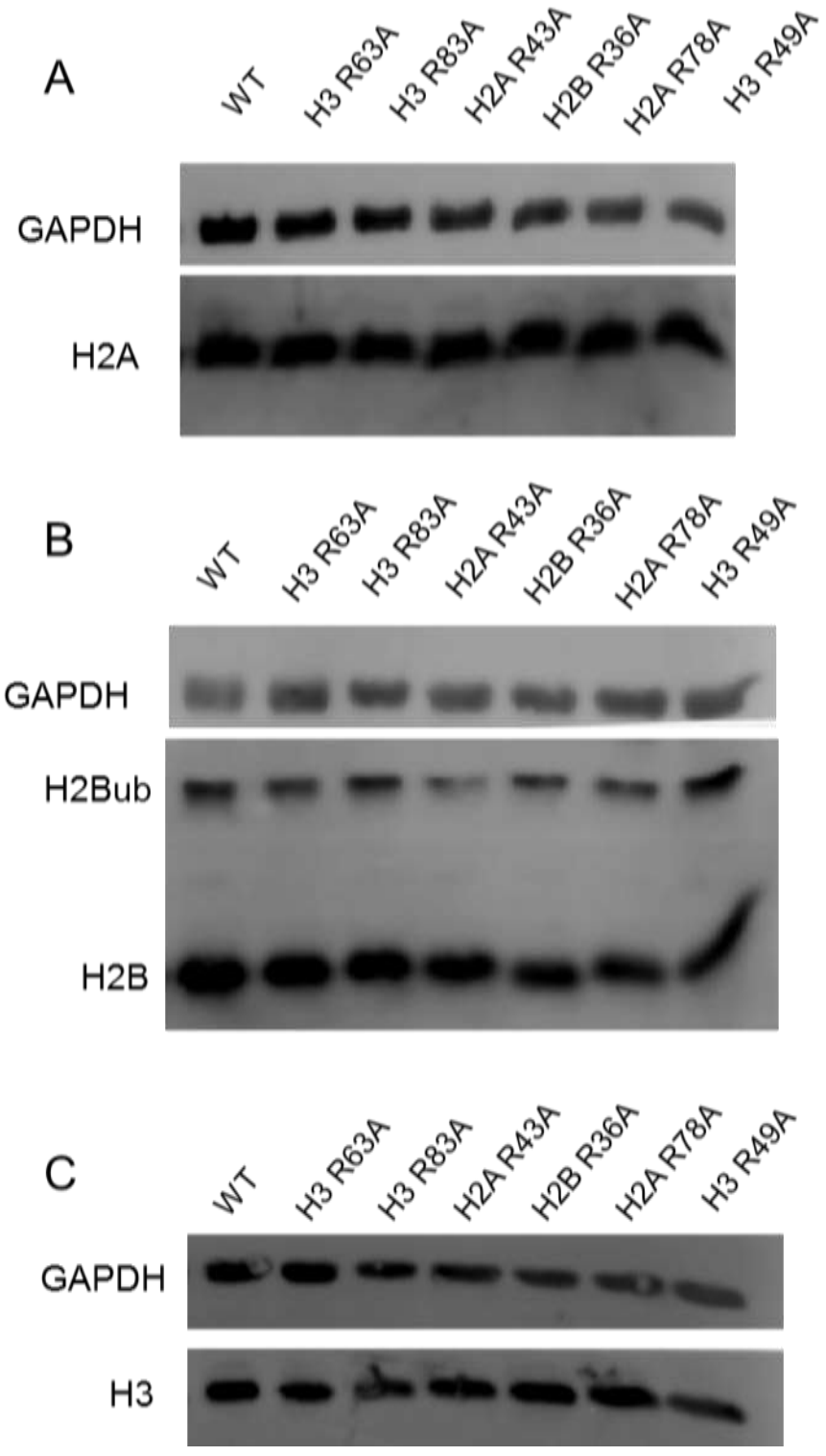


Figure S1. Histone expression levels of viable sprocket arginine mutants. Western blots were probed using antibodies against H2A (A) H2B (B) and H3 (C). GAPDH is used as a loading control.

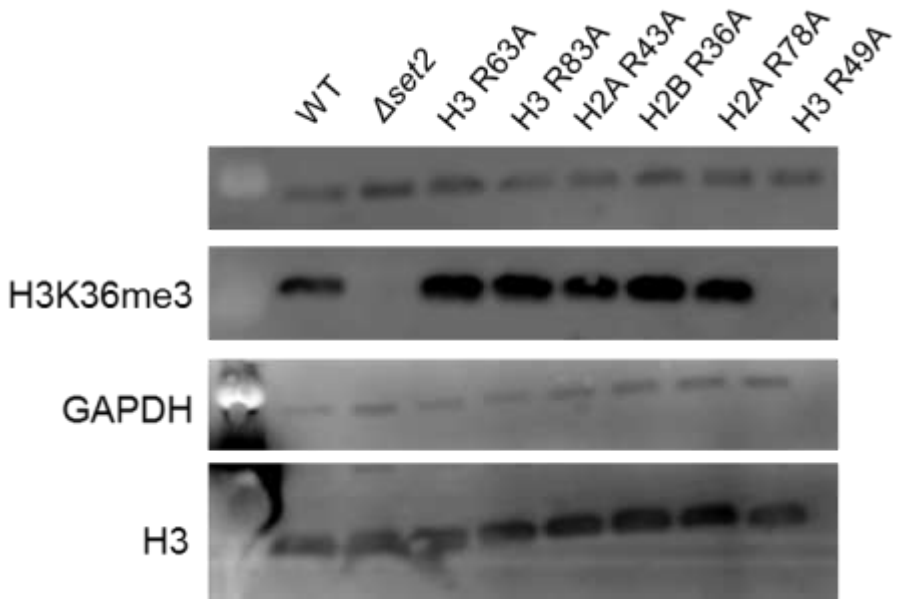


Figure S2. Methylation of H3 K36 in sprocket arginine mutants. Western blots were probed using an antibody specific for H3 K36 trimethylation or for canonical H3. GAPDH is used as a loading control.

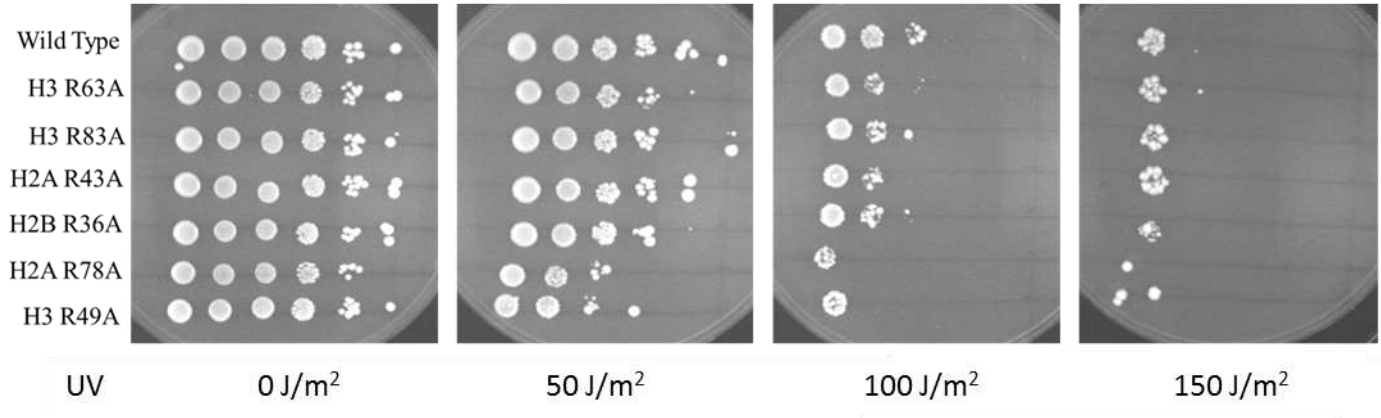
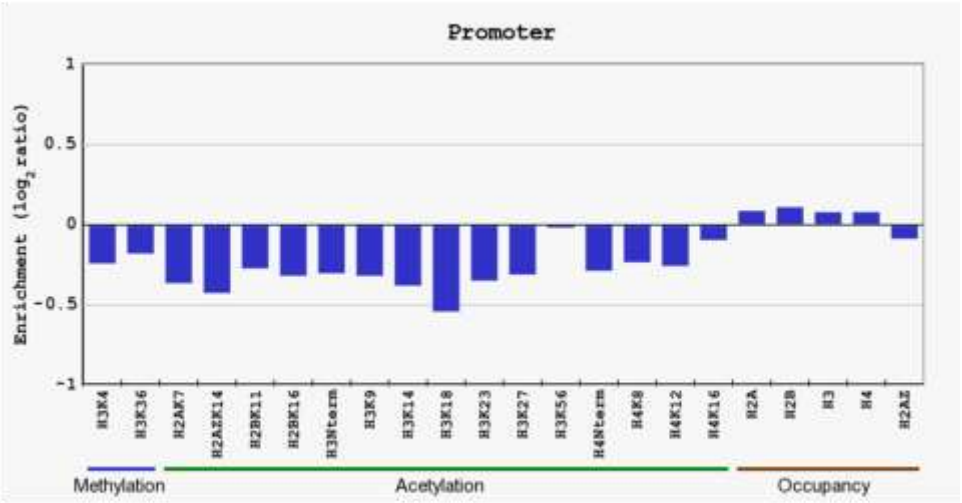


Figure S3. H3 R49A and H2A R78A sprocket arginine mutants are sensitive to UV radiation. Decreasing serial dilutions of mutant yeast cells were spot onto SC plates and exposed to the indicated doses of UV radiation.

A



B

Chromatin Enrichment/Depletion

Promoter				
Histone Modification	Rank Percentile	Trend	P-value	Ranked Gene List
H3K4 Di-Methylation	36.9 %	**Depleted**	**$< 10^{-11}$**	
H3K36 Di-Methylation	39.2 %	**Depleted**	**$< 10^{-7}$**	
H2AK7 Acetylation	26.7 %	**Depleted**	**$< 10^{-12}$**	
H2AZK14 Acetylation	34.6 %	**Depleted**	**$< 10^{-10}$**	
H2BK11 Acetylation	27.6 %	**Depleted**	**$< 10^{-11}$**	
H2BK16 Acetylation	26.5 %	**Depleted**	**$< 10^{-12}$**	
H3Nterm Acetylation	31.0 %	**Depleted**	**$< 10^{-23}$**	
H3K9 Acetylation	29.7 %	**Depleted**	**$< 10^{-9}$**	
H3K14 Acetylation	22.7 %	**Depleted**	**$< 10^{-16}$**	
H3K18 Acetylation	23.4 %	**Depleted**	**$< 10^{-15}$**	
H3K23 Acetylation	26.4 %	**Depleted**	**$< 10^{-12}$**	
H3K27 Acetylation	30.3 %	**Depleted**	**$< 10^{-8}$**	
H3K56 Acetylation	48.6 %	none	0.5017	
H4Nterm Acetylation	29.7 %	**Depleted**	**$< 10^{-26}$**	
H4K8 Acetylation	33.9 %	**Depleted**	**$< 10^{-6}$**	
H4K12 Acetylation	29.5 %	**Depleted**	**$< 10^{-9}$**	
H4K16 Acetylation	41.4 %	none	0.0080	
H2A Occupancy	55.7 %	none	0.0065	
H2B Occupancy	61.8 %	**Enriched**	**$< 10^{-9}$**	
H3 Occupancy	62.2 %	**Enriched**	**$< 10^{-9}$**	
H4 Occupancy	61.1 %	**Enriched**	**$< 10^{-7}$**	
H2AZ Occupancy	44.0 %	none	0.0031	

Figure S4. Genes repressed by the H2A R78 sprocket arginine residue have high levels of promoter histone occupancy and low levels of active histone post-translational modifications. (A) Histone occupancy and histone modification levels in the promoter regions of 290 genes repressed by H2A R78 (i.e., genes with induced expression in the H2A R78A mutant). Analysis was performed on published ChIP-chip data sets using the web-based database ChromatinDB (O'CONNOR AND WYRICK 2007). Note, histone modification data were normalized by nucleosome occupancy levels. (B) Statistical analysis of the data shown in part A showing enrichment or depletion of histone occupancy or post-translational modifications.

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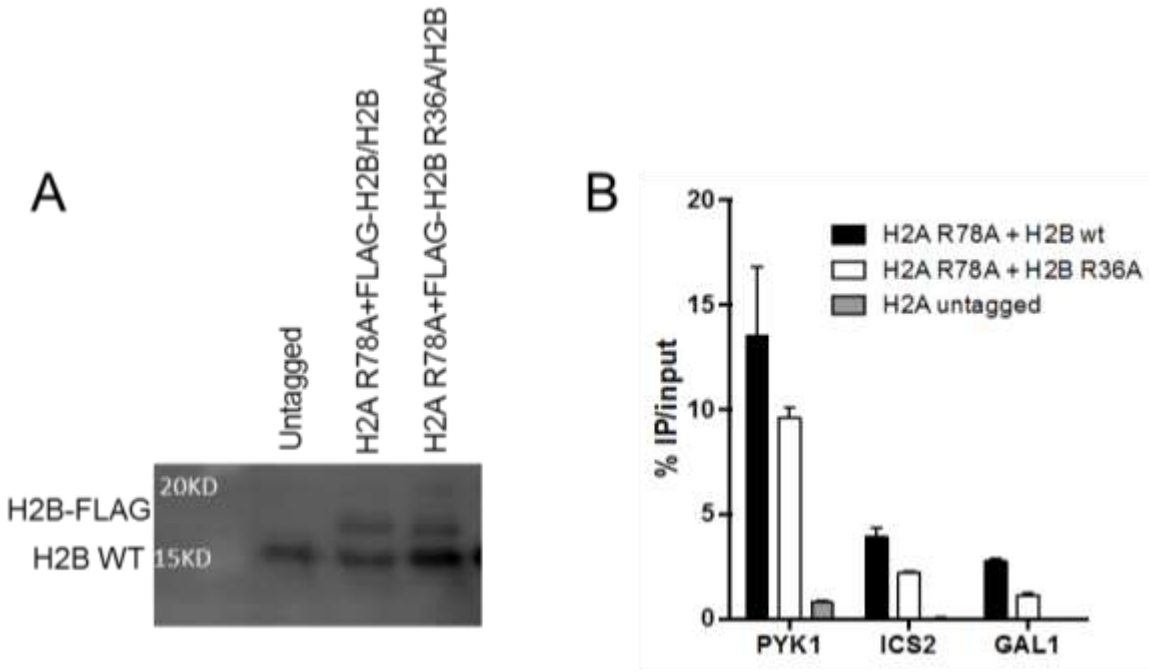

Figure S5. (A) Expression levels of wild type and FLAG-tagged H2B by western blot using an antibody against H2B. Each strain contains an untagged wild type H2B gene as well as the FLAG-tagged H2B. (B) ChIP experiments were conducted as described in Figure 5C.

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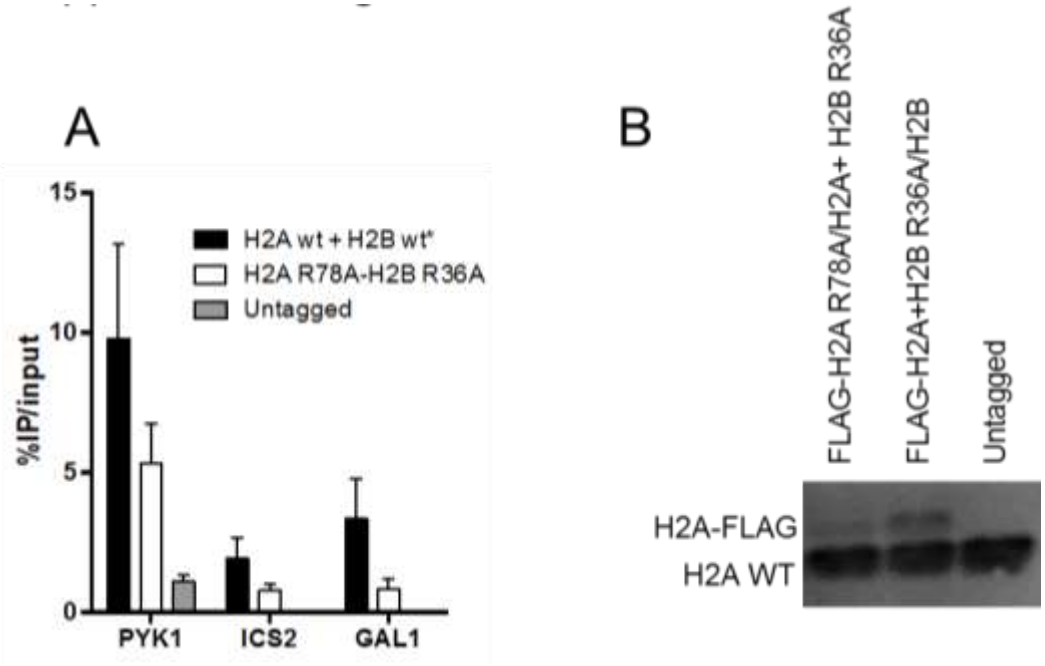

Figure S6. (A) ChIP experiments were conducted as described in Figure 5C, using a FLAG-tagged H2A. Data for the untagged control represents two independent replicates; data for wild type and mutant represents three independent replicates. *H2B wt was heterozygous for H2A R36 (B) Expression levels of wild type and FLAG-tagged H2A by western blot using an antibody against H2A.

TABLE S1. Yeast strains

Strain Name	Genotype	Used in
WY500* (WT)	*MAT**a** his3-1 leu2-0 met15-0 ura3-0 hht1-hhf1::KAN hhf2-hht2::NAT hta1-htb1::HPH hta2-htb2::NAT* p[*CEN URA3 HTA1-HTB1-HHT2-HHF2*]	Fig. 1,5
LT041 (WT snf5Δ HO-LacZ Fusion)	Isogenic to WY500, plus *snf5::HIS3 HO::LacZ*	Fig. 2
LT070 (WT snf5Δ HO-LacZ Fusion)	Isogenic to LT041; WT URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-HHT2-HHF2*]	Fig. 2
LT054 (H4 R45A +WT HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 HTA1-HTB1-HHT2-hhf2_R45A*]	Fig. 2
LT063 (H3 R63A HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 HTA1-HTB1-hht2_R63A-HHF2*]	Fig. 2
LT064 (H3 R83A HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 HTA1-HTB1-hht2_R83A-HHF2*]	Fig. 2
LT061 H2A R43A HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 hta1_R4A3-HTB1-HHT2-HHF2*]	Fig. 2
LT052 H2B R36A HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*]	Fig. 2
LT051 H2A R78A HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*]	Fig. 2
LT062 H3 R49A HO-LacZ Fusion snf5Δ)	Isogenic to LT041 plus p[*CEN LEU2 HTA1-HTB1-hht2_R49A-HHF2*]	Fig. 2
LT060 (WT SNF5 HO-LacZ Fusion)	Isogenic to WY500 plus *HO::LacZ*; WT URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-HHT2-HHF2*]	Fig. 2
LT059 (H4 R45A +WT HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN URA3 HTA1-HTB1-HHT2-HHF2] p*[*CEN LEU2 HTA1-HTB1-HHT2-hhf2_R45A*]	Fig. 2
LT067 H3 R63A HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN LEU2 HTA1-HTB1-hht2_R63A-HHF2]*	Fig. 2
LT068 (H3 R83A HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN LEU2 HTA1-HTB1-hht2_R83A-HHF2]*	Fig. 2
LT065 H2A R43A HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN LEU2 hta1_R43A-HTB1-HHT2-HHF2]*	Fig. 2
LT057 H2B R36A HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*]	Fig. 2

LT056 H2A R78A HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*]	Fig. 2
LT066 H3 R49A HO-LacZ Fusion SNF5)	Isogenic to LT060 plus p[*CEN LEU2 hta1_R78A-HTB1-hht2_R49A-HHF2*]	Fig. 2
WY504 (IJ400) (WT)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-HHT2-HHF2*]	Fig. 2,3,4, S1, S2, S3, S4
YAH034 (WT Flo8-His3 Fusion)	Isogenic to WY500 plus *FLO8::HIS3;* URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-HHT2-HHF2*]	Fig. 2
YEW2 (H4 R45H Flo8-HIS3 Fusion)	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-HHT2-hhf2_R45H*]	Fig. 2
YAH035 (H3 R63A Flo8-HIS3 Fusion	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-hht2_R63A-HHF2*]	Fig. 2
YAH036 (H3 R83A Flo8-HIS3 Fusion)	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-hht2_R83A-HHF2*]	Fig. 2
YAH037 (H2A R43A Flo8-HIS3 Fusion	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 hta1_R43A-HTB1-HHT2-HHF2*]	Fig. 2
YAH038 (H2B R36A Flo8-HIS3 Fusion)	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*]	Fig. 2
YAH039 (H2A R78A Flo8-HIS3 Fusion)	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*]	Fig. 2
YAH040 (H3 R49A Flo8-HIS3 Fusion)	Isogenic to YAH034; URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-hht2_R49A-HHF2*]	Fig. 2
LT019 (H3 R49A)	Isogenic to WY500; URA replaced with p[*CEN LEU2 HTA1-HTB1-hht2_R49A-HHF2*]	Fig. 3,4, S1, S2, S3
LT023 (H2A R78A)	Isogenic to WY500; URA replaced with p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*]	Fig. 3,4, S1, S2, S3, S4
LT015 (H3 R63A)	Isogenic to WY500; URA replaced with p[*CEN LEU2 HTA1-HTB1-hht2_R63A-HHF2*]	Fig. 4, S1, S2, S3
LT016 (H3 R83A)	Isogenic to WY500; URA replaced with p[*CEN LEU2 HTA1-HTB1-hht2_R83A-HHF2*]	Fig. 4, S1, S2, S3
LT017 (H2A R43A)	Isogenic to WY500; URA replaced with p[*CEN LEU2 hta1_R43A-HTB1-HHT2-HHF2*]	Fig. 4, S1, S2, S3
LT018 (H2B R36A)	Isogenic to WY500; URA replaced with p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*]	Fig. 4, S1, S2, S3

WY502 (*set2Δ*)	Isogenic to WY500 plus *set2Δ::*KAN	Fig. S2
RM001.1 (*mag1Δ*)	Isogenic to WY504 plus *mag1Δ::HIS3*	Fig. 4
WY471.1 (WT Gal Shutdown ΔL1)	Isogenic to WY500; URA plasmid replaced with p[*CEN HIS3 HTA1-pGAL-HTB1-HHT2-HHF2*] p[*CEN LEU2 hta1_Δ39-42-HTB1-HHT2-HHF2*]	Fig. 5
WY471.3 (WT Gal Shutdown Leu2)	Isogenic to WY500; URA plasmid replaced with p[*CEN HIS3 HTA1-pGAL-HTB1-HHT2-HHF2*] p[*CEN LEU2*]	Fig. 5
WY471.18 (WT Gal Shutdown H2B R36A)	Isogenic to WY500; URA plasmid replaced with p[*CEN HIS3 HTA1-pGAL-HTB1-HHT2-HHF2*] p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*]	Fig. 5
WY471.23 (WT Gal Shutdown H2A R78A)	Isogenic to WY500; URA plasmid replaced with p[*CEN HIS3 HTA1-pGAL-HTB1-HHT2-HHF2*] p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*]	Fig. 5
WY471.31 (WT Gal Shutdown H2A R78A-H2B R36A)	Isogenic to WY500; URA plasmid replaced with p[*CEN HIS3 HTA1-pGAL-HTB1-HHT2-HHF2*] p[*CEN LEU2 hta1_R78A-htb1_R36A-HHT2-HHF2*]	Fig. 5
YAH031 (FLAG-tagged WT)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*] p[*CEN HIS3 HTA1-FLAGtag_htb1-HHT2-HHF2*]	Fig. 5
YAH011 (WT untagged)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*] p[*CEN HIS3 hta1_R78A-HTB1-HHT2-HHF2*]	Fig. 5, S5
YAH012 (FLAG-tagged H2B-H2A R78A)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*] p[*CEN HIS3 hta1_R78A-FLAGtag_htb1-HHT2-HHF2*]	Fig. 5, S5
YAH013 (FLAG-tagged H2B R36A-H2A R78A)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*] p[*CEN HIS3 hta1_R78A-FLAGtag-htb1_R36A-HHT2-HHF2*]	Fig. 5, S5
YT09 (WT untagged)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 HTA1-HTB1-HHT2-HHF2*] p[*CEN URA3 HTA1-HTB1-HHT2-HHF2*]	Fig. S6
YT011 (FLAG-tagged H2A R78A H2B R36A)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*] p[*CEN URA3 FLAGtag_hta1_R78A-htb1_R36A-HHT2-HHF2*]	Fig. S6
YT012 (H2A FLAG-tagged WT*)	Isogenic to WY500; URA plasmid replaced with p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*] p[*CEN URA3 FLAGtag_HTA1-HTB1-HHT2-HHF2*]	Fig. S6

*Strain WY500 is isogenic to JHY205 (AHN *et al*. 2005)

TABLE S2. Plasmids transformed into WY500 for spotting assays

Plasmid Name	**Genotype**
pJW500* (WT)	p[*CEN LEU2 HTA1-HTB1-HHT2-HHF2*]
pLT15 (H3 R63A)	p[*CEN LEU2 HTA1-HTB1-hht2_R63A-HHF2*]
pLT16 (H3 R83A)	p[*CEN LEU2 HTA1-HTB1-hht2_R83A-HHF2*]
pLT17 (H2A R43A)	p[*CEN LEU2 hta1_R43A-HTB1-HHT2-HHF2*]
pLT18 (H2B R36A)	p[*CEN LEU2 HTA1-htb1_R36A-HHT2-HHF2*]
pLT23 (H2A R78A)	p[*CEN LEU2 hta1_R78A-HTB1-HHT2-HHF2*]
pLT19 (H3 R49A)	p[*CEN LEU2 HTA1-HTB1-hht2_R49A-HHF2*]
pLT20 (H3 R63A + H3 R83A)	p[*CEN LEU2 HTA1-HTB1-hht2_R63A_R83A-HHF2*]
pLT21 (H3 R83 + H2A R43A)	p[*CEN LEU2 hta1_R43A-HTB1-hht2_R83A-HHF2*]
pLT22 (H2A R43A + H2B R36A)	p[*CEN LEU2 hta1_R43A-htb1_R36A-HHT2-HHF2*]
pLT24 (H2A R78A + H3 R49A)	p[*CEN LEU2 hta1_R78A-HTB1-hht2_R49A-HHF2*]
pLT31 (H2A R78A + H2B R36A)	p[*CEN LEU2 hta1_R78A-htb1_R36A-HHT2-HHF2*]

*Plasmid pJW500 is identical to pQQ18 (AHN *et al*. 2005)

TABLE S3. Microarray data for H2A R78A mutant

Gene Name	Fold Change
CCL1	N.S.*
KIN28	N.S.
RAD1	N.S.
RAD2	N.S.
RAD3	N.S.
RAD4	1.3
RAD7	N.S.
RAD10	N.S.
RAD14	N.S.
RAD16	1.4
RAD23	N.S.
RAD26	N.S.
RAD28	N.S.
SSL1	N.S.
SSL2	N.S.
TFB1	N.S.
TFB2	N.S.
TFB3	N.S.
TFB4	N.S.
TFB5	N.S.

*N.S.: Not Significant