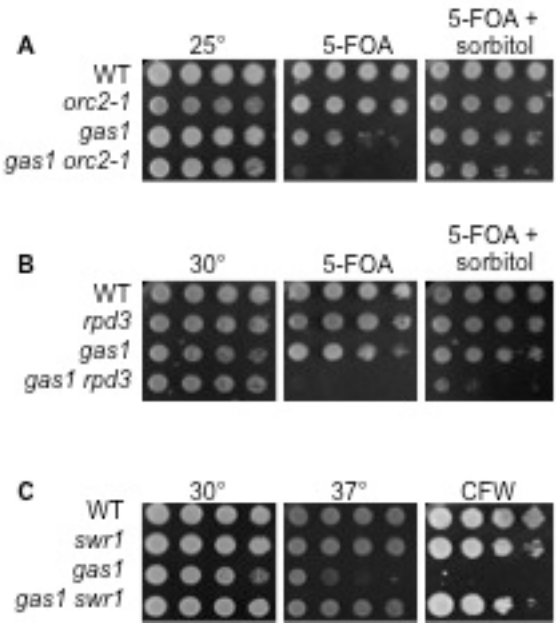

Figure S1 The synthetic lethality of *gas1* with *orc2-1* or *rpd3* is at least partially rescued by sorbitol, whereas deletion of *SWR1* rescued both *gas1* temperature and CFW sensitivities. (A) Wild type (LPY10266), *orc2-1* (LPY10267), *gas1* (LPY10271) and *gas1 orc2-1* covered by p-*GAS1* (LPY10270) were plated on SC or SC with 5-FOA, to counterselect p-*GAS1, URA3*, with or without 1M sorbitol at 25°. (B) Wild type (LPY4196), *rpd3* (LPY14355), *gas1* (LPY19200), *gas1 rpd3* covered by p-*GAS1, URA3* (LPY15695) were plated at 30° on SC and SC with 5-FOA, to counterselect p-*GAS1, URA3*, with or without 1M sorbitol. (C) Wild type (LPY5), *swr1* (LPY16104), *gas1* (LPY10129) and *gas1 swr1* (LPY17161) were plated on SC at 30°, 37°, and SC with CFW at 30°.

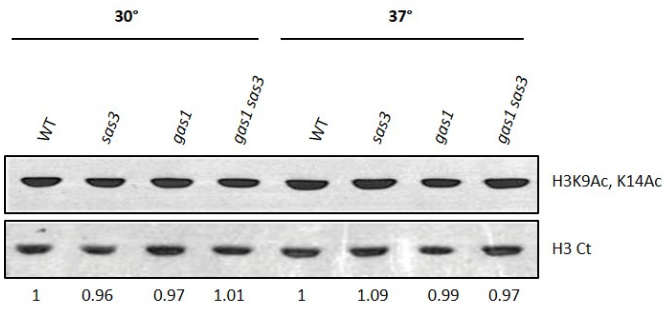

Figure S2 Neither *gas1* nor *gas1 sas3* have significantly reduced global levels of H3K9Ac, K14Ac. Whole cell lysates from wild type (LPY5), *sas3* (LPY8256), *gas1* (LPY10129) and *gas1 sas3* (LPY17520) were separated on 18% SDS-PAGE after growth at either 30° or 37° and probed with anti-H3K9Ac, K14Ac (1:10000; Millipore). Blots were reprobed with anti-H3 C-terminal (Ct) (1:10000; Millipore) as a loading control.

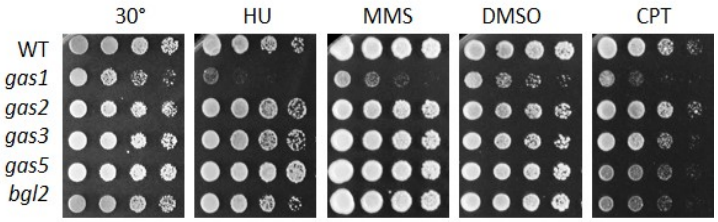

Figure S3 Genotoxin sensitivity is not a common feature of the *GAS* family or cell wall disruption. Wild type (LPY5), *gas1* (LPY10129), *gas2* (LPY10047), *gas3* (LPY10051), *gas5* (LPY11544) and *bgl2* (LPY13102) were plated on SC or SC with HU, MMS or CPT, with DMSO as a control, and incubated at 30°. Among the five-membered *GAS* family, *GAS2*, like *GAS4* (not shown) is expressed meiotically, whereas *GAS1*, *GAS3*, and *GAS5* are vegetatively expressed (Ragni *et al.* 2007). *BGL2* encodes a cell wall endo-β-1,3-glucanase (Mrsa *et al.* 1993).

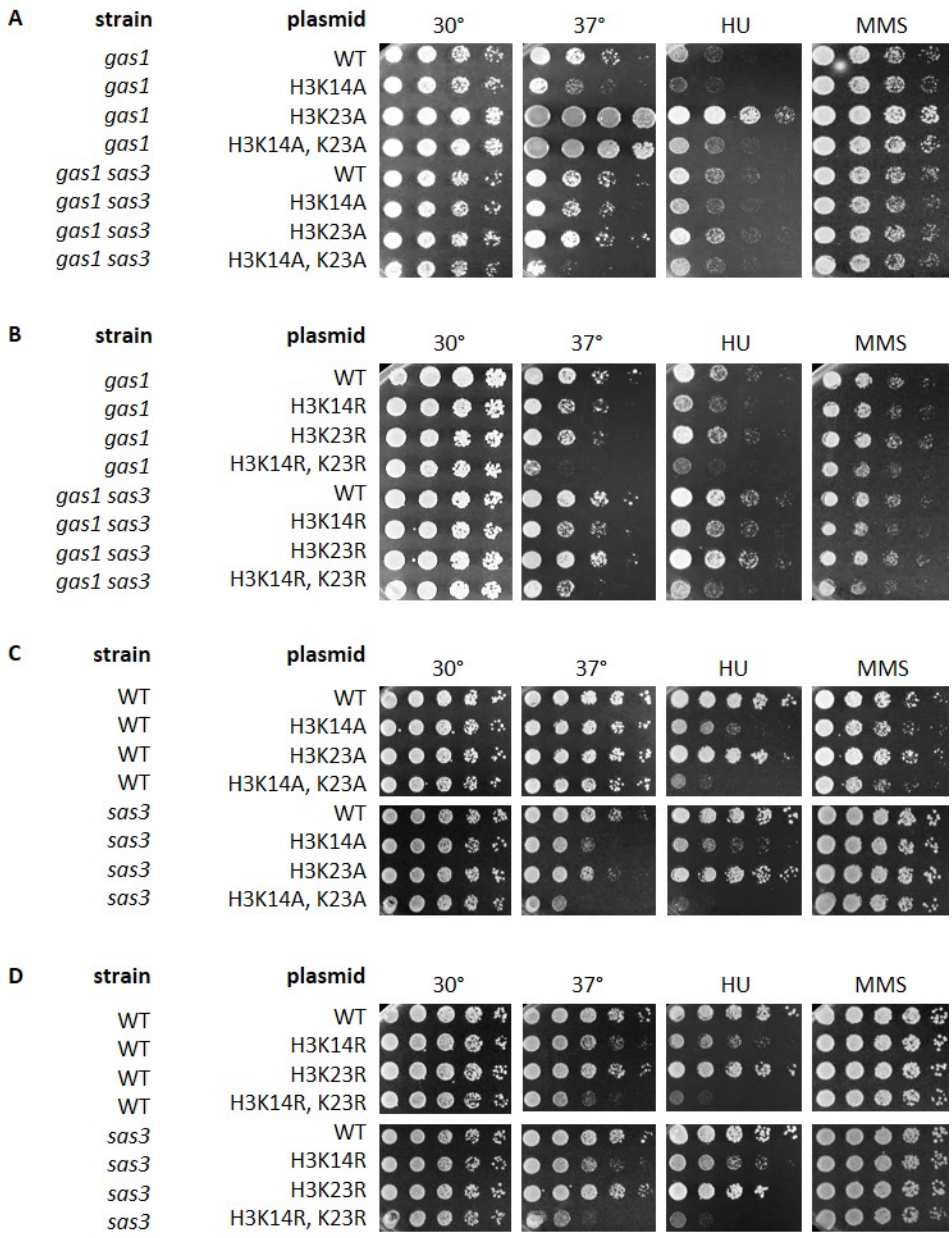

Figure S4 H3K23A mutants suppress *gas1* temperature and DNA damage sensitivity phenotypes. (A) H3K23A mutant in *gas1* rescues temperature, HU and MMS sensitivity. This suppression is decreased in the absence of *SAS3* as well as in the double mutant H3K14A, K23A. (B) Mutation of the same single residues to arginine does not alter phenotypes of either *gas1* or *gas1 sas3* yet, as in A, the double mutant exacerbates the phenotypes. (C/D) Wild type and *sas3* controls analyzed as in A and B. Although phenotypes are similar to wild type, *sas3* decreased growth at elevated temperature. For these experiments *gas1* (LPY18343), *gas1 sas3* (LPY19878), wild type (LPY12242) and *sas3* (LPY16432) were freshly transformed with indicated histone mutants and struck out on 5-FOA to select against the covering wild type plasmid (pJH33; Ahn *et al.* 2005). Transformations were performed with plasmids containing wild type H3-H4 (*HHT2-HHF2*; pLP1775), H3K14A (pLP1777), H3K23A (pLP3086), H3K14A, K23A (pLP3078), H3K14R (pLP3018), H3K23R (pLP3050) and H3K14R, K23R (pLP3064). Mutants were generated with site-directed mutagenesis with oligonucleotides listed in Table S3.

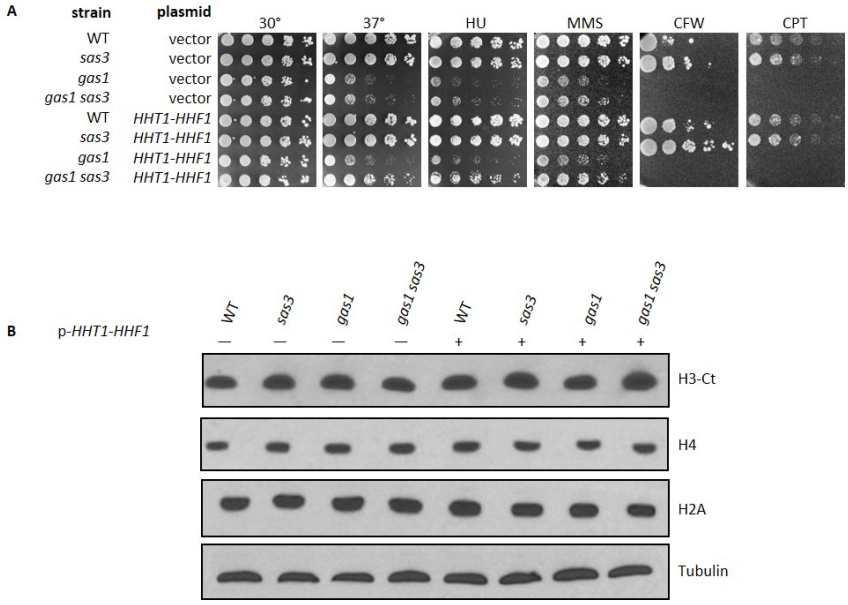

Figure S5 Suppression of *gas1* phenotypes by deletion of *SAS3* is at least partially dependent on the presence of *HHT1-HHF1.* (A) Diminished suppression by deletion of *SAS3* is observed in the histone mutant background deleted for *HHT1-HHF1*. Suppression is restored when the *HHT1-HHF1* locus is provided on a CEN plasmid in the *gas1 sas3* double mutant. (B) However, this is not due to global changes in histone levels. Genotoxin and growth conditions are the same as in Figure S4. Strains are as in Figure S4, except those carrying the p-*HHT1-HHF1* (pLP3145), which also have *HHT2-HHF2* (pLP1775). Strains plated in (A) were subsequently used for analysis in (B). The immunoblot was probed with anti-H3-Ct (1:10000; Millipore), anti-H4 (1:10000; Millipore), anti-H2A (1:5000; Abcam) and anti-tubulin (1:10000; Bond *et al.* 1986) as a loading control.

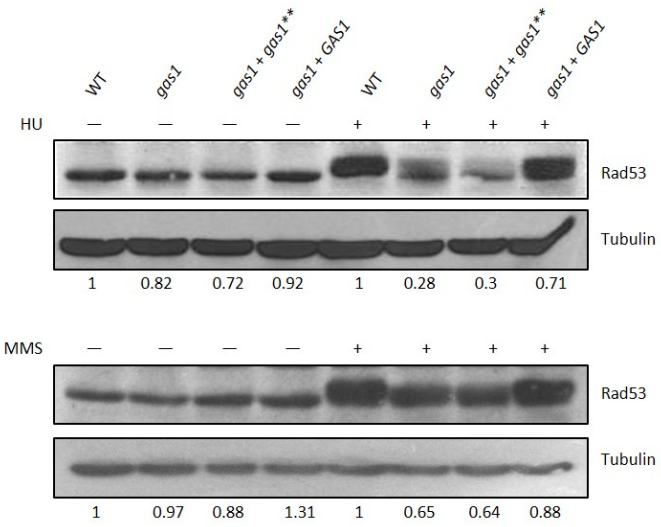

Figure S6 Reduction of Rad53 protein levels and phosphorylation isoforms is dependent on the β-1,3-glucanosyltransferase activity of Gas1. Wild type (LPY5), *gas1* (LPY10129), *gas1* + p-*gas1*** (LPY12251) and *gas1* + p-*GAS1* (LPY122326) were treated with HU or MMS. Whole cell lysates were separated on 8% SDS-PAGE and probed with anti-Rad53 followed by anti-tubulin as loading control, as done for Figure 5.

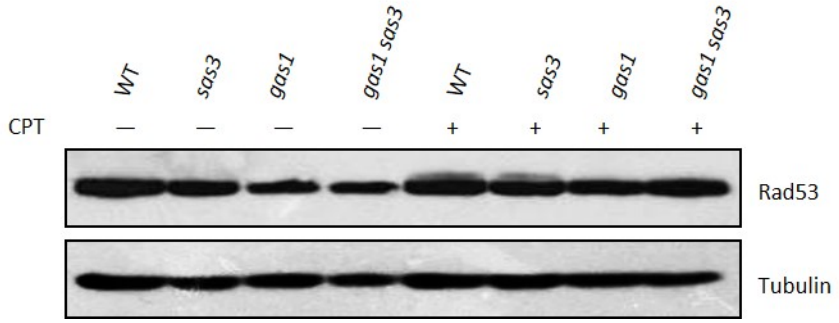

Figure S7 Rad53 is only minimally, if at all, phosphorylated following exposure to CPT as previously reported (Redon *et al.* 2003). Strains and treatment are the same as in Figure 5.

Table S1 Yeast strains used in this study

Strain	Genotype	Source
LPY5 (W303-1a)	*MATα ade2-1 can1-100 his3-11,15 leu2-3,112 trp1-1 ura3-1*	R. Rothstein
LPY1597	W303 *MATa sas2Δ::TRP1*	
LPY2444	*MATα his3Δ200 leu2Δ1 ura3-52* with rDNA Ty mURA insert	J.S. Smith
LPY2447	*MATα his3Δ200 leu2Δ1 ura3-52* with rDNA Ty mURA insert *sir2Δ2::HIS3*	J.S. Smith
LPY4196	LPY5 + pLP60	
LPY4924	W303 *MATa hmr::TRP1* TELVR::*URA3*	
LPY5035	W303 *MATa sir2Δ::HIS3 hmr::TRP1* TELVR::*URA3*	
LPY5526	W303 *MATa yng1Δ::HIS3 rDNA::ADE2-CAN1 TELVR::URA3*	
LPY6285	W303 *MATa rDNA::ADE2-CAN1 TELVR::URA3*	K. Runge
LPY6439	W303 *MATa ada2Δ::kanMX*	R. Rothstein
LPY8242	W303 *MATa gcn5Δ::HIS3*	
LPY8256	W303 *MATa sas3Δ::HIS3*	
LPY9820	W303 *MATa gas1Δ::kanMX rDNA::ADE2-CAN1 TELVR::URA3*	
LPY10047	W303 *MATa rDNA::ADE2-CAN1 hmr::TRP1 gas2Δ::kanMX*	
LPY10051	W303 *MATa rDNA::ADE2-CAN1 hmr::TRP1 gas3Δ::kanMX*	
LPY10074	*MATα his3Δ200 leu2Δ1 ura3-52* with rDNA Ty mURA insert *gas1Δ::kanMX*	
LPY10129	*MATa ade2-1 can1-100 his3-11,15 leu2-3,112 trp1-1 ura3-1 gas1Δ::kanMX*	
LPY10266	W303 *MATα rDNA::CAN:ADE2 +* pLP1823	
LPY10267	W303 *MATα orc2-1 rDNA::CAN:ADE2 +* pLP1823	
LPY10270	W303 *MATα gas1Δ::kanMX orc2-1 rDNA::CAN:ADE2 +* pLP1823	
LPY10271	W303 *MATα gas1Δ::kanMX orc2-1 rDNA::CAN:ADE2 +* pLP1823	
LPY11544	W303 *MATa gas3Δ::kanMX rDNA::CAN:ADE2 hmr::TRP1*	
LPY12232	W303 *MATa hht1-hhf1Δ::kanMX hht2-hhf2Δ::kanMX hta2-htb2Δ::HPH* + pJH33	M.M. Smith
LPY12247	LPY10129 *+* pLP135	
LPY12251	LPY10129 *+* pLP2114	
LPY12264	W303 *MATa rDNA::CAN:ADE2 hmr::TRP1 gcn5Δ::NatMX*	
LPY12326	LPY10129 + pLP1951	
LPY13102	W303 *MATa rDNA::CAN:ADE2 hmr::TRP1 bgl2Δ::kanMX*	
LPY14355	W303 *MATa rpd3Δ::kanMX +* pLP60	
LPY15695	W303 *MATa gas1Δ::kanMX rpd3Δ::kanMX +* pLP60 + pLP1823	
LPY16039	W303 *MATa sas3Δ::HIS3*	
LPY16104	W303 *MATa swr1Δ::kanMX*	
LPY16432	W303 *MATa hht1-hhf1Δ::kanMX hht2-hhf2Δ::kanMX hta2-htb2Δ::HPH sas3Δ::HIS3* + pJH33	
LPY16444	W303 *MATa sas3Δ::HIS3 hmr::TRP1* TELVR::*URA3*	
LPY16736	W303 *MATa gas1Δ::kanMX gcn5Δ::HIS3 +* pLP1640	
LPY16798	W303 *MATa gas1Δ::kanMX gcn5Δ::HIS3* + pLP 1640 + pLP 135	
LPY16800	W303 *MATa gas1Δ::kanMX gcn5Δ::HIS3* + pLP 1640 + pLP 1950	

LPY16801	W303 *MATa gas1Δ::kanMX gcn5Δ::HIS3* + pLP 1640 + pLP 2114	
LPY16914	W303 *MATa spt20Δ::HIS3*	D. Stillman
LPY16997	W303 *MATa gas1Δ::kanMX yng1Δ::HIS3 rDNA::ADE2-CAN1 TELVR::URA3*	
LPY17161	*W303 MATa swr1Δ::kanMX gas1Δ::kanMX* TELVR*::URA3*	
LPY17370	W303 *MATa ahc1Δ::kanMX*	
LPY17685	*MATa his3Δ200 leu2Δ1 ura3-52* with rDNA Ty mURA insert *gas1Δ::kanMX sas3Δ::HIS3*	
LPY18050	LPY5 + pLP 135	
LPY18081	LPY10129 + pLP 135	
LPY18206	W303 *MATa rtg2Δ::kanMX*	
LPY18343	W303 *MATa hht1-hhf1Δ::kanMX hht2-hhf2Δ::kanMX hta2-htb2Δ::HPH gas1Δ::kanMX* + pJH33	
LPY18372	W303 *MATa gas1Δ::kanMX rtg2Δ::kanMX*	
LPY18518	W303 *MATa ahc2Δ::kanMX*	
LPY19101	W303 *MATa gas1Δ::kanMX gcn5Δ::HIS3 sas3Δ::HIS3* + pLP 1640	
LPY19200	LPY10129 *+* plp60	
LPY19272	W303 *MATa gas1Δ::kanMX ada2Δ::kanMX*	
LPY19414	W303 *MATa gas1Δ::kanMX ahc2Δ::kanMX*	
LPY19467	W303 *MATa gas1Δ::kanMX ahc1Δ::kanMX*	
LPY19630	W303 *MATa gas1Δ::kanMX spt20Δ::HIS3*	
LPY19670	W303 *MATa gas1Δ::kanMX sas2Δ::HIS3*	
LPY19731	W303 *MATa sas3Δ::HIS3 hmr::TRP1* TELVR::*URA3*	
LPY19771	W303 *MATa gas1Δ::kanMX sgf73Δ::URA3*	
LPY19773	W303 *MATa gas1Δ::kanMX hmr::TRP1* TELVR::*URA3*	
LPY19816	W303 *MATa sgf73Δ::URA3*	
LPY19823	W303 *MATa gas1Δ::kanMX sas3Δ::HIS3*	
LPY19878	W303 *MATa hht1-hhf1Δ::kanMX hht2-hhf2Δ::kanMX hta2-htb2Δ::HPH sas3Δ::HIS3 gas1Δ::kanMX* + pJH33	

All strains were constructed during the course of this study or are part of our standard lab collection unless otherwise indicated.
All strains are W303 unless otherwise indicated.

Table S2 Plasmids used in this study

Plasmid	Description	Alias	Source
pJH33	*HTA1 HTB1 HHF2 HHT2 URA3* CEN		Ahn *et al.* 2005
pLP60	vector HIS3 CEN	pRS313	
pLP135	vector *LEU2* 2μ	YEP351	
pLP1640	*GCN5 URA3* CEN		S. Lo
pLP1775	*HHT2-HHF2 TRP1* CEN		S.L. Berger
pLP1777	*hht2-K14A HHF2 TRP1* CEN		S. Lo
pLP1823	vector *TRP1* 2μ	pRS424	C. Nislow
pLP1950	*gcn5-KQL LEU2* 2μ		
pLP1951	*GAS1 LEU2* 2μ		
pLP2114	*gas1-E161Q, E262Q LEU2* 2μ		
pLP3018	*hht2-K14R HHF2 TRP1* CEN		
pLP3050	*hht2-K23R HHF2 TRP1* CEN		
pLP3064	*hht2-K14R, K23R HHF2 TRP1* CEN		
pLP3078	*hht2-K14A, K23A HHF2 TRP1* CEN		
pLP3086	*hht2-K23A HHF2 TRP1* CEN		
pLP3145	*HHT1-HHF1 URA3* CEN		

All plasmids were constructed during the course of this study or are part of our standard lab collection unless otherwise indicated.

Table S3 Oligonucleotides used in this study

Oligo #	Sequence	Name
oLP1965	CCA CTG GTG GTA GAG CCC CAA G	H3K14R sense
oLP1966	CTT GGG GCT CTA CCA CCA GTG G	H3K14R antisense
oLP1969	CAA TTA GCC TCC AGG GCT GCC AG	H3K23R sense
oLP1970	CTG GCA GCC CTG GAG GCT AAT TG	H3K23R antisense
oLP1985	CAA TTA GCC TCC GCG GCT GCC AG	H3K23A sense
oLP1986	CTG GCA GCC GCG GAG GCT AAT TG	H3K23A antisense

Supplemental Literature Cited

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Mrsa, V., F. Klebl, and W. Tanner, 1993 Purification and characterization of the *Saccharomyces cerevisiae BGL2* gene product, a cell wall endo-beta-1,3-glucanase. J. Bacteriol. 175: 2102-2106.

Ragni, E., T. Fontaine, C. Gissi, J.P. Latge, and L. Popolo, 2007 The Gas family of proteins in *Saccharomyces cerevisiae*: characterization and evolutionary analysis. Yeast 24: 297-308.