Figure S1

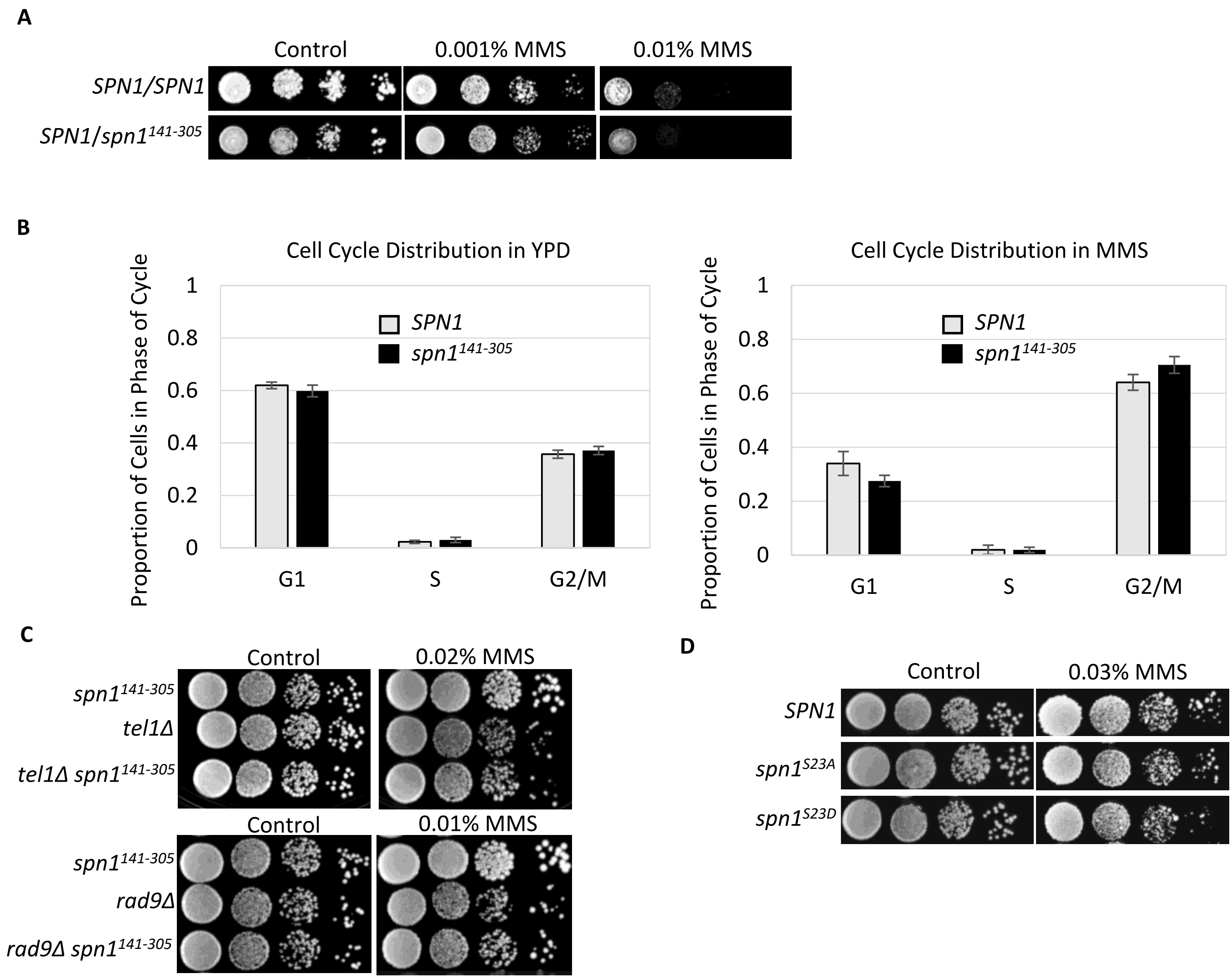

Figure S1. Further examination of effects of MMS exposure on strains expressing Spn1 mutants. A) Ten-fold serial dilutions of cells expressing endogenous Spn1 and plasmid-borne Spn1 (*SPN1/SPN1*) or Spn1$^{141\text{-}305}$ (*SPN1/spn1*$^{141\text{-}305}$). Cells were grown on SC-His plates containing increasing concentrations of MMS. B) To determine cell cycle distribution by budding index, logarithmically growing cells in YPD (left panel) or YPD containing 0.03% MMS (right panel) were fixed and examined by microscopy. C) Ten-fold serial dilutions of the *spn1*$^{141\text{-}305}$, *tel1Δ, tel1Δspn1*$^{141\text{-}305}$*, rad9Δ* and *rad9Δspn1*$^{141\text{-}305}$ strains were grown on YPD and YPD plus MMS plates. Deletion of Tel1 or Rad9 results in strain sensitivity when grown on MMS (CHANG *et al*. 2002; KAPITZKY *et al*. 2010) D) Ten-fold serial dilutions of the *SPN1, spn1*^{S23A} and *spn1*^{S23D} strains were grown on YPD and YPD plus MMS plates.

Figure S2

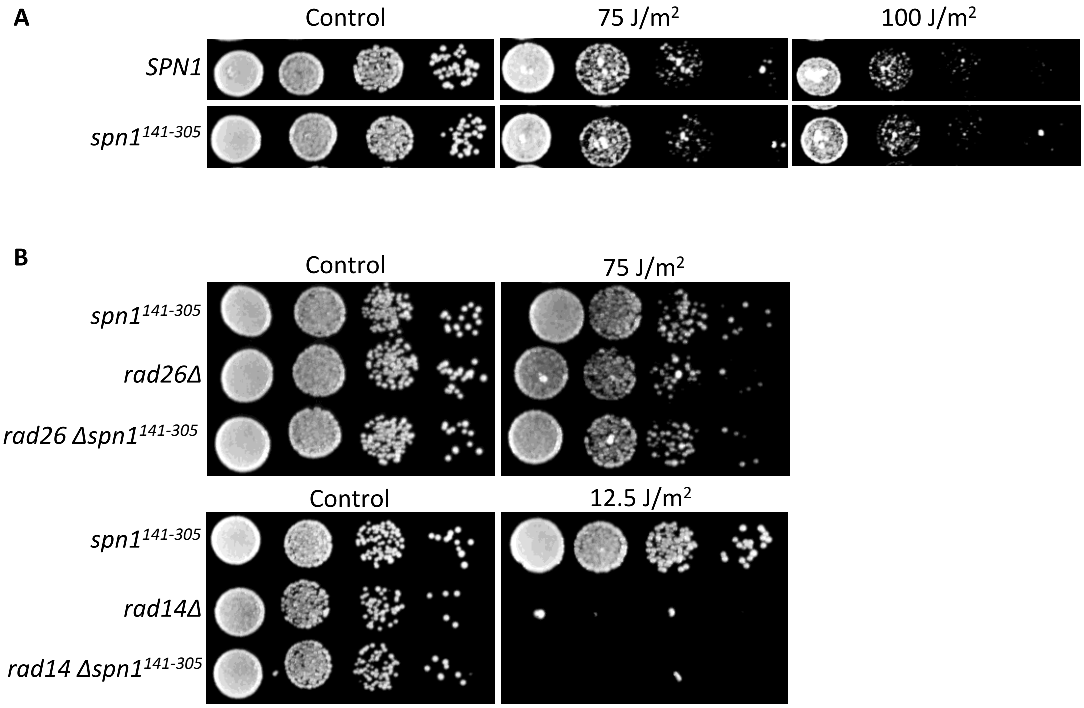

Figure S2. Growth phenotypes after exposure to UV. A) Ten-fold serial dilutions of cells expressing Spn1 or Spn1$^{141\text{-}305}$ were spotted onto YPD plates and exposed to increasing doses of UV. B) Ten-fold serial dilutions of cells expressing Spn1 or Spn1$^{141\text{-}305}$ in the in *rad26Δ* and *rad14Δ* backgrounds were spotted onto YPD plates and exposed to UV**.** Strains are: *SPN1, spn1*$^{141\text{-}305}$*, rad26Δ, rad26Δspn1*$^{141\text{-}305}$*, rad14Δ, and rad14Δspn1*$^{141\text{-}305}$.

Figure S3

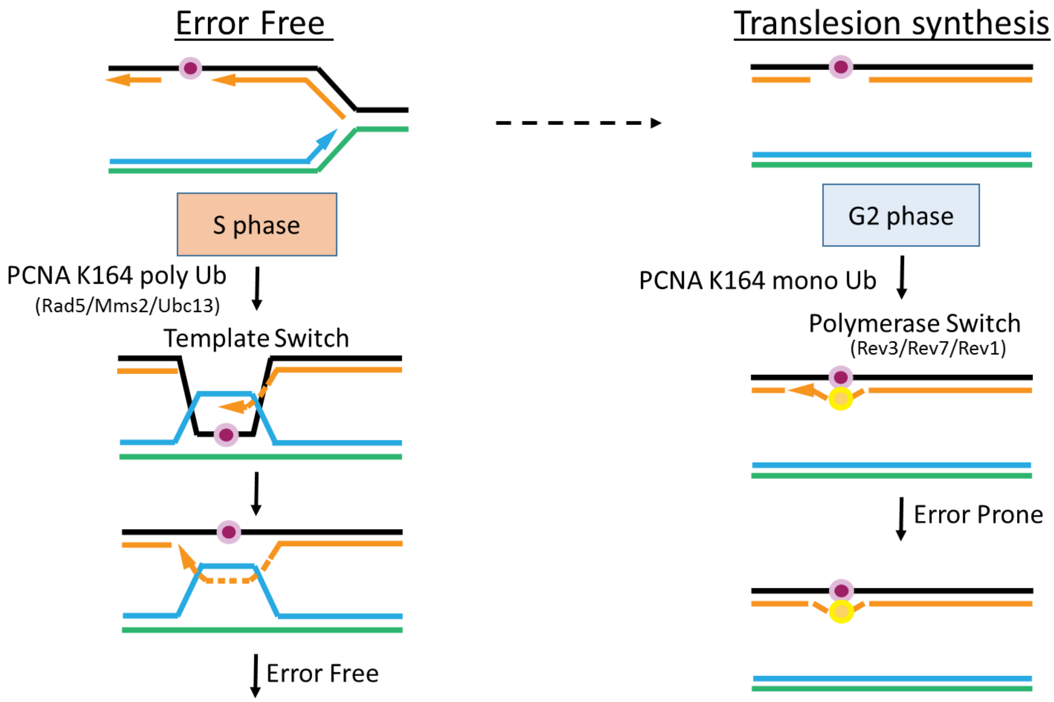

Figure S3. The sub-pathways of DNA Damage Tolerance (DDT). Diagram depicting the DDT pathway. The DDT pathway consists of two branches, error free and translesion synthesis. Image adapted from (BRANZEI and PSAKHYE 2016).

Figure S4

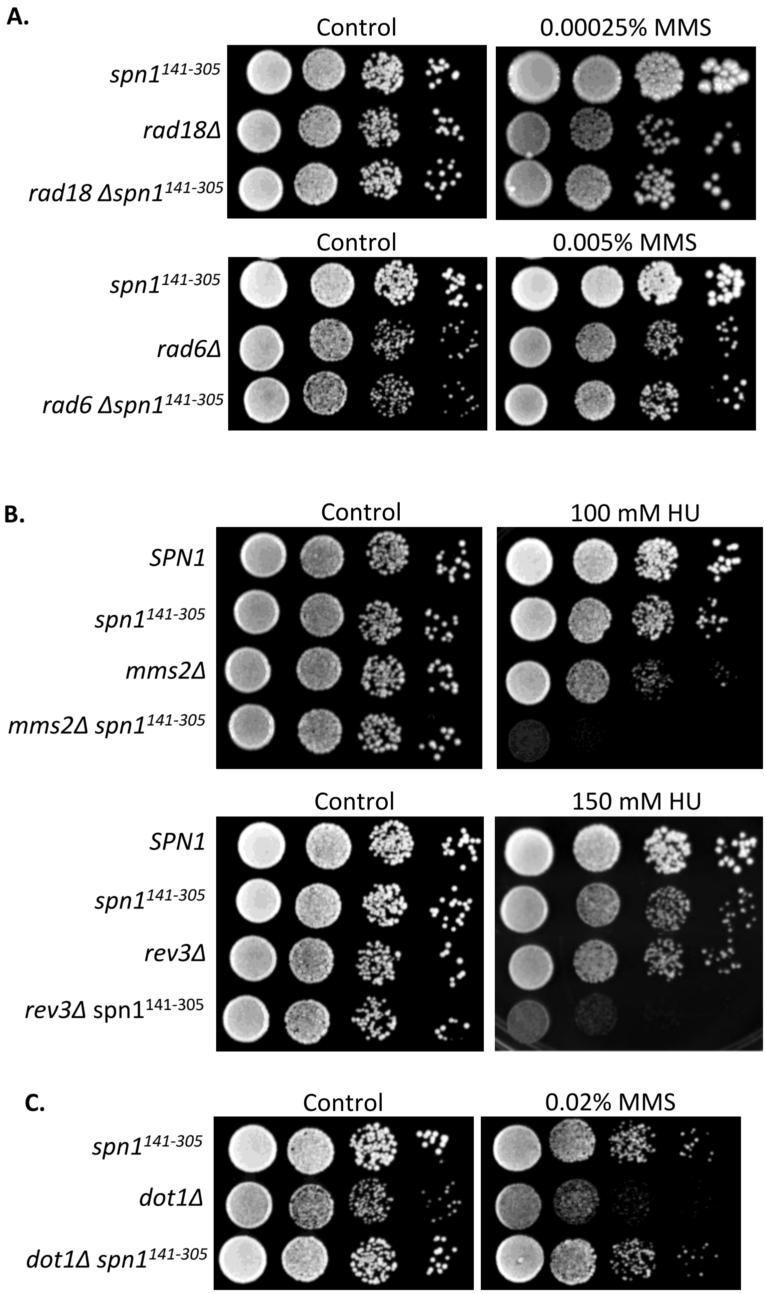

Figure S4. Growth phenotypes of *SPN1* and the DDT pathway. A) Ten-fold serial dilutions of cells expressing Spn1 or Spn1$^{141-305}$ in the *rad18Δ* and *rad6Δ* strains. Cells were grown on YPD and YPD plus MMS plates. Strains are: *spn1*$^{141-305}$, *rad18Δ, rad18Δspn1*$^{141-305}$, *rad6Δ,* and *rad6Δspn1*$^{141-305}$. B) Ten-fold serial dilutions of cells expressing Spn1 or Spn1$^{141-305}$ in the *mms2Δ and rev3Δ strains*. Strains were grown on YPD and YPD plates containing hydroxyurea (HU). Strains are: *SPN1*, *spn1*$^{141-305}$, *mms2Δ, mms2Δspn1*$^{141-305}$, *rev3Δ, and rev3Δspn1*$^{141-305.}$ C) Ten-fold serial dilutions of *spn1*$^{141-305}$, *dot1Δ* and *dot1Δspn1*$^{141-305}$strains. Cells were grown on YPD and YPD plus MMS plates.

Figure S5

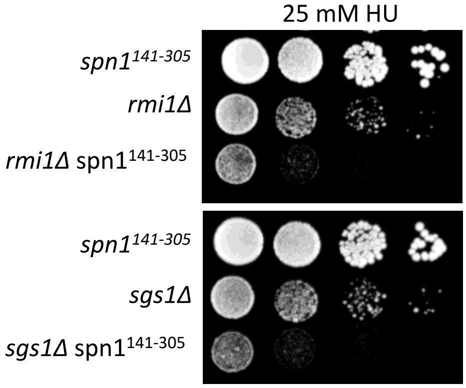

Figure S5. Expression of Spn1$^{141\text{-}305}$ in *rmi1Δ* and *sgs1Δ* strains enhances cell sensitivity in HU. Ten-fold serial dilutions of *spn1*$^{141\text{-}305}$, *rmi1Δ, rmi1Δspn1*$^{141\text{-}305}$, *sgs1Δ* and *sgs1Δspn1*$^{141\text{-}305}$ strains. Cells were grown on YPD plus HU plates.

TABLE S1. STRAINS

Name	**Description**	**Genotype**	**Source**
BY4741	BY4741	MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0	Thermo Scientific
LZ1	*SPN1*	BY4741 + *spn1Δ::LEU2*, pCR311	Li *et al.*, Zhang *et al.*
LZ3	*spn1*$^{141-305}$	BY4741 + *spn1Δ::LEU2*, pCR344	Li *et al.,* Zhang *et al.*
AT241	*spn1*^{S23A}	BY4741 + *spn1Δ::LEU2*, pAT101	This study
AT242	*spn1*^{S23D}	BY4741 + *spn1Δ::LEU2*, pAT102	This study
AT252	*SPN1SPN1*	BY4741 + pCR311	This study
AT254	*SPN1spn1*$^{141-305}$	BY4741 + pCR344	This study
AT102	*apn1ΔSPN1*	*apn1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT104	*apn1Δspn1*$^{141-305}$	*apn1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT122	*dot1ΔSPN1*	*dot1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT124	*dot1Δspn1*$^{141-305}$	*dot1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT138	*mag1ΔSPN1*	*mag1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT140	*mag1Δspn1*$^{141-305}$	*Mag1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT142	*mms2ΔSPN1*	*mms2Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT144	*mms2Δspn1*$^{141-305}$	*mms2Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT182	*rad5ΔSPN1*	*rad5Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT184	*rad5Δspn1*$^{141-305}$	*rad5Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
CR58A	*rad6ΔSPN1*	*rad6Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
CR58C	*rad6Δspn1*$^{141-305}$	*rad6Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
CR60A	*rad9ΔSPN1*	*rad9Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
CR60C	*rad9Δspn1*$^{141-305}$	*rad9Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT162	*rad14ΔSPN1*	*rad14Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT164	*rad14Δspn1*$^{141-305}$	*rad14Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT166	*rad18ΔSPN1*	*rad18Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT168	*rad18Δspn1*$^{141-305}$	*rad18Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT174	*rad26ΔSPN1*	*rad26Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT176	*rad26Δspn1*$^{141-305}$	*rad26Δ::KanMX+ spn1Δ::LEU2*, pCR344	This study
AT186	*rad51ΔSPN1*	*rad51Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT188	*rad51Δspn1*$^{141-305}$	*rad51Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT194	*rad55ΔSPN1*	*rad55Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT196	*rad55Δspn1*$^{141-305}$	*rad55Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT198	*rad57ΔSPN1*	*rad57Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT200	*rad57Δspn1*$^{141-305}$	*rad57Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT202	*rev1ΔSPN1*	*rev1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT204	*rev1Δspn1*$^{141-305}$	*rev1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT206	*rev3ΔSPN1*	*rev3Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT208	*rev3Δspn1*$^{141-305}$	*rev3Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT210	*rev7ΔSPN1*	*rev7Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT212	*rev7Δspn1*$^{141-305}$	*rev7Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT214	*rmi1ΔSPN1*	*rmi1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT216	*rmi1Δspn1*$^{141-305}$	*rmi1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
CR82A	*sgs1ΔSPN1*	*sgs1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
CR82C	*sgs1Δspn1*$^{141-305}$	*sgs1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT230	*tel1ΔSPN1*	*tel1Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT232	*tel1Δspn1*$^{141-305}$	*tel1Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study
AT238	*ubc13ΔSPN1*	*ubc13Δ::KanMX* + *spn1Δ::LEU2*, pCR311	This study
AT240	*ubc13Δspn1*$^{141-305}$	*ubc13Δ::KanMX* + *spn1Δ::LEU2*, pCR344	This study

TABLE S2. PLASMIDS

Plasmids	Description
pCR 311	Full length wild type *SPN1* with 403 bp of upstream sequence and 116bp of downstream sequence, myc2 tagged at the amino terminus, pRS313 (CEN, *HIS3*)
pAA 344	*spn1*$^{141\text{-}305}$ with 403 bp of upstream sequence and 116bp of downstream sequence, myc2 tagged at the amino terminus, pRS313 (CEN, *HIS3*)
pAT 101	*spn1*^{S23A} with 403 bp of upstream sequence and 116bp of downstream sequence, myc2 tagged at the amino terminus, pRS313 (CEN, *HIS3*)
pAT 102	*spn1*^{S23D} with 403 bp of upstream sequence and 116bp of downstream sequence, myc2 tagged at the amino terminus, pRS313 (CEN, *HIS3*)

TABLE S3. DNA PRIMERS

Name	Primer Sequence	Description
STA763	GTTTATAGTTGACTTTTGGGCGGAAGCTGTCCCATCTTC	Spn1 S23A reverse
STA764	GAAGATGGGACAGCTTCCGCCCAAAAGTCAACTATAAAC	Spn1 S23A forward
STA765	CGTTTATAGTTGACTTTTGGTCGGAAGCTGTCCCATCTTCTG	Spn1 S23D reverse
STA766	CAGAAGATGGGACAGCTTCCGACCAAAAGTCAACTATAAACG	Spn1 S23D forward

Supplemental References:

BRANZEI, D., and I. PSAKHYE, 2016 DNA damage tolerance. Curr Opin Cell Biol **40:** 137-144.

CHANG, M., M. BELLAOUI, C. BOONE and G. W. BROWN, 2002 A genome-wide screen for methyl methanesulfonate-sensitive mutants reveals genes required for S phase progression in the presence of DNA damage. Proc Natl Acad Sci U S A **99:** 16934-16939.

KAPITZKY, L., P. BELTRAO, T. J. BERENS, N. GASSNER, C. S. ZHOU *et al.*, 2010 Cross-species chemogenomic profiling reveals evolutionarily conserved drug mode of action. Molecular Systems Biology **6**.

LI, S., A. R. ALMEIDA, C. A. RADEBAUGH, L. ZHANG, X. CHEN *et al.*, 2018 The elongation factor Spn1 is a multi-functional chromatin binding protein. Nucleic Acids Res **46:** 2321-2334.

ZHANG, L., A. G. FLETCHER, V. CHEUNG, F. WINSTON and L. A. STARGELL, 2008 Spn1 regulates the recruitment of Spt6 and the Swi/Snf complex during transcriptional activation by RNA polymerase II. Mol Cell Biol **28:** 1393-1403.