

Supplementary Table 2 - List of plasmids used in this study

Plasmid	Protein Expressed
p976	pGex-6P-2-GST
p677	pET21a-Net1(1-600)-His ₆
p700	pET21b-GST-Cdc14
p782	pET15b-His ₆ -Cdc14 (1-374)
p1289	pET15b-His ₆ -GFP-Cdc14
p1290	pET15b-His ₆ -1HA-Cdc14
p1293	pET15b-His ₆ -GFP-Cdc14-Q106L
p1294	pET15b-His ₆ -GFP-Cdc14-W108R
p1295	pET15b-His ₆ -Cdc14 (1-374)
p1297	pET15b-His ₆ -GFP-Cdc14-GETS (V120G, D121E, P122T, P123S)
p1328	pET21b-GST-Cdc14-Tab6 (P116L)
p1330	pET21b-GST-Cdc14-W108R

Supplementary Table 3 - List of peptides used in this study

Peptide	N-terminus	Sequence	C-terminus	Figures
Chk1 - WT	acetyl	FTDVPALNYPATPPPH	amide	Fig. 1, 3, Sup. Fig. 6
Chk1 - P86G	acetyl	FTDVGALNYPATPPPH	amide	Fig. 1
Sic1 - WT	acetyl	PSTTKSFKNAPLLAPP	amide	Fig. 1, 3
Sic1 - P56G	acetyl	PSTTKSFKNAGLLAPP	amide	Fig. 1
Kar9 - WT	acetyl	KGKLLSSVPPLPYDE	amide	Fig. 1
Kar9 - P508G	acetyl	KGKLLSSVGPLPYDE	amide	Fig. 1
Chk1-L88G	acetyl	FTDVPAGNYPATPPPH	amide	Sup. Fig. 1
Chk1-F82G, V85G	acetyl	GTDGPALNYPATPPPH	amide	Sup. Fig. 1
Chk1-Y90G	acetyl	FTDVPALNGPATPPPH	amide	Sup. Fig. 1
Chk1-Δ91-97	acetyl	FTDVPALNY	amide	Sup. Fig. 1
biotin - Chk1 - WT	biotin - aminohexanoic acid	FTDVPALNYPATPPPH	amide	Fig. 4, Sup. Fig. 2, 9
biotin - Chk1 - scr	biotin - aminohexanoic acid	YVPAPAPPTTPLNFDHP	amide	Fig. 4, Sup. Fig. 2, 9
PxL - Sic1 disordered + SPxAAA	acetyl	FTDVPALNYPATPPPH	amide	Fig. 6
AxG - Sic1 disordered + SPxAAA	acetyl	FTDVAAGNYPATPPPH-TSPLG(p)SPLAAFNGLTSPQRSFPFKSSVK	amide	Fig. 6
PxL - Sic1 disordered + TPxAAA	acetyl	FTDVPALNYPATPPPH-TSPLG(p)TPLAAFNGLTSPQRSFPFKSSVK	amide	Sup. Fig. 9
AxG - Sic1 disordered + TPxAAA	acetyl	FTDVAAGNYPATPPPH-TSPLG(p)TPLAAFNGLTSPQRSFPFKSSVK	amide	Sup. Fig. 9

Supplementary Table 4 - List of yeast strains used in this study

Strain name	Genotype	Background
1262	<i>MATa cdc14-1</i>	W303
4636	<i>MATa cdc14-1, trp1::TRP1-CDC14pr-CDC14-GFP</i>	W303
5488	<i>MATa CBK1-HA6::HIS3</i>	W303
5489	<i>MATa CBK1-HA6::HIS3 CDC14-MYC18::TRP1</i>	W303
5493	<i>MATa CBK1-AxG (P86A, L88G)-HA6::HIS3</i>	W303
5494	<i>MATa CBK1-AxG (P86A, L88G)-HA6::HIS3 CDC14-MYC18::TRP1</i>	W303
5498	<i>MATa CBK1-HA6::HIS3 hof1Δ::TRP1</i>	W303
5506	<i>MATa CBK1-AxG (P86A, L88G)-HA6::HIS3 hof1Δ::TRP1</i>	W303
5507	<i>MATa CBK1-HA6::HIS3 cyk3Δ::TRP1</i>	W303
5508	<i>MATa CBK1-AxG (P86A, L88G)-HA6::HIS3 cyk3Δ::TRP1</i>	W303
5511	<i>MATa prGAL1-CDC20::LEU2 ORC6-PxL (FTDVPALNYPAAPPPH)-PK3::TRP1 FIN1-HA3::URA3</i>	W303
5515	<i>MATa prGAL1-CDC20::LEU2 ORC6-PK3::TRP1 FIN1-HA3::URA3</i>	W303
5519	<i>MATa cdc14-1 trp1::TRP1-pCDC14-CDC14-GFP CBK1-HA6::HIS3</i>	W303
5520	<i>MATa cdc14-1 trp1::TRP1-pCDC14-CDC14-W108R-GFP CBK1-HA6::HIS3</i>	W303
5521	<i>MATa prGAL1-CDC20::LEU2 ORC6-PxL (FTDVPALNYPAAPPPH)-PK3::TRP1 FIN1-HA3::URA3 CDC14-MYC18::HIS3</i>	W303
5522	<i>MATa prGAL1-CDC20::LEU2 ORC6-PK3::TRP1 FIN1-HA3::URA3 CDC14-MYC18::HIS3</i>	W303
5523	<i>MATa cdc14-1 trp1::TRP1-pCDC14-CDC14-GETS (V120G, D121E, P122T, P123S)-GFP</i>	W303