

Appendix for:

PLK1 inhibition delays mitotic entry revealing changes to the phosphoproteome of mammalian cells early in division

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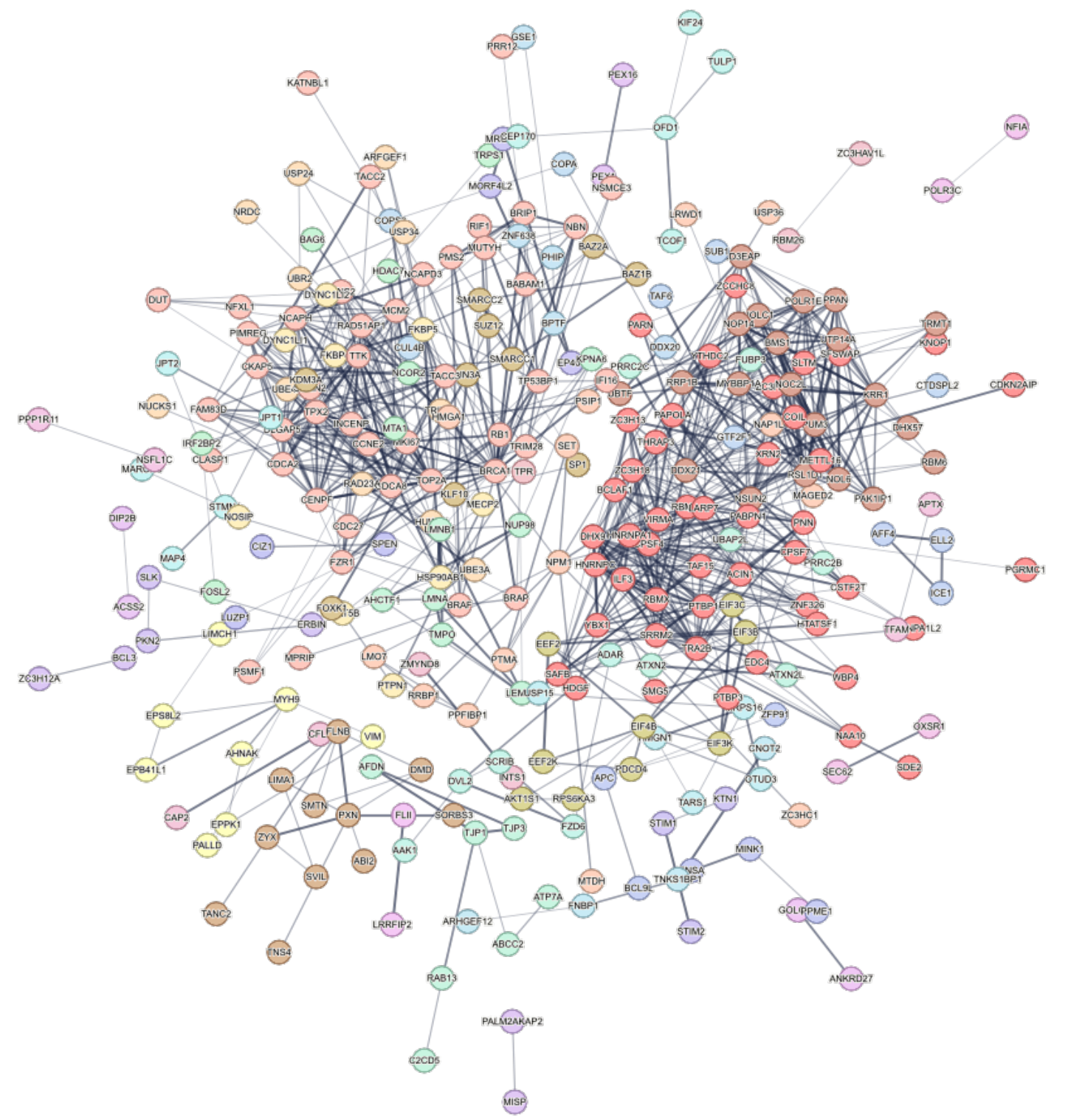
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Appendix Figure S1.



Appendix Figure S1:

Network generated by STRING database analysis depicting differentially phosphorylated proteins specific to prolonged prophase after applying linear modeling. The gene names of the corresponding proteins are shown.

Appendix Table S1.

Gene name	Protein name	Position	Sequence window	Log2 Fold Change	Adjusted p value
BABAM1	BRISC and BRCA1-A complex member 1	29	EPRPRTSRNPEGAED	-0.13564	0.452216
		65	ADDGSLNTSGAGPKS	3.204965	0.016133
BRAF	Serine/threonine-protein kinase B-raf	429	PQRERKSSSSSEDRN	0.020957	0.950661
		732	HRSASEPSLNRAFGQ	-0.46754	0.021532
BRAP	BRCA1-associated protein	97	ERKSSEASPTAQRSK	0.758572	0.036624
		571	IAMASASSPASSGGS	1.233362	0.15329
BRCA1	Breast cancer type 1 susceptibility protein	1524	LQNRNYPQQEELIKV	-1.30686	0.007531
		967	GNETGLITPNKHGLL	-0.17774	0.826994
BRIP1	Fanconi anemia group J protein	1032	SSENSASSPPRFKTE	0.035918	0.946208
		226	CSTKQGNSEQSSNTI	-1.83983	0.001085
CCNE2	G1/S-specific cyclin-E2	990	IVISRSTPTFNKQT	-0.23038	0.223636
		21	PQPSQTESPQEAQII	-2.4767	0.000111
CDC27	Cell division cycle protein 27 homolog	220	NRLNLESSNSKYSLN	-1.35576	0.004893
CDC42	Cell division cycle-associated protein 2	710	EPKAGTDSVPVCASV	1.182501	0.000399
CDCA8	Borealin	219	YNISGNGSPPLADSKE	1.338073	0.001152
		154	PSKKRTQSIQKGKGG	0.468087	0.752849
CENPF	Centromere protein F	821	CRLEADQSPKNSAIL	0.501219	0.225936
		2922	SVTEKRLSSGQNKAS	0.573635	0.094435
		1121	MTDNQNNKSKEAGGL	3.917572	0.001717
		3079	VNNLPERSPPTDSPRE	2.153058	0.000459
		3054	KVKVAQRSPVDSGTI	0.114245	0.720904
CKAP5	Cytoskeleton-associated protein 5	144	VSLNPCNTPQKIFTT	0.911767	0.011247
		816	FEKMQGQSPAPATRG	1.182614	0.044594
CLASP1	CLIP-associating protein 1	1223	SRDGGAAASPATEGRG	0.340762	0.124086
		1091	SSNTSVGSPSNTIGR	1.696642	0.040077
DLGAP5	Disks large-associated protein 5	646	RIRTRRQSSGSATNV	0.307958	0.44256
		656	SATNVASTPDNRRGS	1.075414	0.101246
DUT	Deoxyuridine 5'-triphosphate nucleotidohydrolase, mitochondrial	662	GIPQQTITSPENAGPQ	2.025834	0.000199
		777	KNTASQNSILEEGET	0.407237	0.715857
FAM64A	Protein FAM64A	248	RGSGGFGSTGKN	1.260963	0.002084
FAM83D	Protein FAM83D	106	RIQESQSGTKWLV	1.82903	0.009454
		16	GTSVRRRLQHQEQL	0.592557	0.057232
FZRL1	Fizzy-related protein homolog	462	GTQSTEGSPVSKMSV	1.241782	0.001824
GIN52	DNA replication complex GINS protein PSF2	70	RINENEKSPSQNRKA	0.825274	0.00045
IFI16	Gamma-interferon-inducible protein 16	182	QPLESTQSQDF	-1.61545	0.001327
		106	KKEVDATSPAPSTSS	0.976518	0.002982
INCENP	Inner centromere protein	153	EEQTQPPSPAGAGMS	-0.22222	0.496304
		123	KTEGAEATPGAQKRK	-0.25215	0.559768
		446	GPREPPQSAARRKRSY	-0.0091	0.988528
		263	KLRIAQVSPGPRDSP	0.986793	0.150242
KATNBL1	KATNB1-like protein 1	421	NPKPAASSPETPSAG	1.649486	0.031907
		481	PRSKTPSPSPCASKV	0.066519	0.826603
MCM2	DNA replication licensing factor MCM2	135	SGSSQTESPSKYSYG	0.816669	0.007063
		13	ESFTMASSPAQRRRG	-0.20928	0.503571
MKI67	Antigen KI-67	139	RRGLIYDSDEEDEER	-0.19919	0.461216
		27	GNDPLTSSPGRSSRR	-1.03073	0.026971
		584	VISPPAPSPRKTPVA	0.204023	0.909901
		2783	PAASVTGSRRRRPAP	1.784411	0.031297
		1253	TTKIPCDSPQSDPVD	1.950414	0.00924
		1329	LTENLTGSKRRPQTP	0.812784	0.592089
		1815	QPGNLPGSNNRLQTR	-0.13797	0.787722
		2022	VGKLTQITSGKTTQTH	1.108794	0.020309
		357	QYSQQQNSPQKHKNK	-0.04028	0.939795
		1376	TKMPCESSPPESADT	1.319096	0.005691
		374	YTTGRRESVNLGKSE	-0.17389	0.739664
		648	MICKRRSGASEANL	1.680698	0.24483
		621	PKRGGGRKSGNLPSKR	1.771414	0.027648
		1628	DPDKNPASSKRRLKT	0.515792	0.524267
		328	RDVESVQTPSKAVGA	0.014021	0.984762
		2773	LKESAKQTPAPAAASV	0.50022	0.7761
		1327	LDLTENLTGSKRRPQ	0.454921	0.213038
MPRIIP	Myosin phosphatase Rho-interacting protein	1503	SQDPDVPDTPTSKPKQ	0.396177	0.455166
MUTYH	A/G-specific adenine DNA glycosylase	891	GGGEATGSLAQGKD	1.011246	0.001912
NBN	Nibrin	512	SKRSQVSSPCKSRKP	0.420998	0.023535
NCAPD3	Condensin-2 complex subunit D3	343	TTPGPSLSQGVSVDE	-1.53502	0.000459
NCAPH	Condensin complex subunit 2	517	FSYQRQTSNRSEPSG	-1.86704	0.02044
		15	LPATMNNSSSETRGH	2.490922	0.032082
NDNL2	Melanoma-associated antigen G1	459	KQDAPSQSENKKKST	0.078348	0.943523
		87	DSPRLLASPSSRSID	0.861254	0.38164
NFXL1	NF-X1-type zinc finger protein NFXL1	304	PSGPAAPS	1.001316	0.008161
		64	SQG5QGPSPPQGARRA	1.064173	0.029366
PMS2;PMS2CL	Mismatch repair endonuclease PMS2;Protein PMS2CL	50	GAVPSGTSPGCVATT	1.595657	0.033303
PRR12	Proline-rich protein 12	403	MVEKQDQSPSLRTGE	1.193663	0.00045
PSMF1	Proteasome inhibitor P131 subunit	917	AGAYRSPQQTAKAP	-1.10341	0.015456
RAD51AP1	RAD51-associated protein 1	153	WEKANVSSPHREFPP	1.074455	0.003203
		120	VTTNVQNSQDKSIEK	-1.65658	0.004118
RB1	Retinoblastoma-associated protein	327	LVVSVKSPNQSLRL	0.582974	0.183504
		249	AVIPINGSRTPRRG	0.77095	0.007063
RIF1	Telomere-associated protein RIF1	356	DSFETQRTPRKSNLD	0.240314	0.15906
		2161	SLVSANDSPSGMQTR	0.736809	0.003275
		1579	WKNKSNEVDIQDQE	0.03069	0.925885
		1688	KRLHKRDSFDNCSLG	-0.41527	0.187504
SLAIN2	SLAIN motif-containing protein 2	2195	RSQEDEISSPVNKKVR	0.319566	0.418184
		2196	SQEDEISSPVNKKVRR	0.215008	0.539879
TACC2	Transforming acidic coiled-coil-containing protein 2	63	IPSSGAASPRGFPLG	1.578614	0.000635
		48	AGSLGPGSPVRAGAS	0.000119	0.999812
TACC3	Transforming acidic coiled-coil-containing protein 3	1635	PRSTCAPSPQREVLT	1.402082	0.008431
		2256	DSGGQEDSPAKGLSV	0.519993	0.036132
TOP2A	DNA topoisomerase 2-alpha	2226	GSGRRVQNSPPVGRKT	-0.17854	0.215832
		317	AGRAMTILSPQEEVAA	0.369546	0.423235
TP53BP1	Tumor suppressor p53-binding protein 1	434	SEAQPPESPETRLGQ	0.822245	0.001201
		1106	DEEENEESDNEKETE	-0.05939	0.862834
		1247	KNENTEGSPQEDGVE	0.102269	0.858992
		4	MEVSPLOQPVNE	0.940392	0.003275
TPX2	Targeting protein for Xklp2	1213	QMAEVLPSPRGQRVI	1.924142	7.36E-05
		862	TQEKTSNLSITEDSKM	1.845217	0.104161
TPX2	Targeting protein for Xklp2	552	IEDTEPMSPVLSNKF	-2.26502	0.003153
		1028	AVAESVASPQKTMVSV	-0.50497	0.007574
TRIM28	Transcription intermediary factor 1-beta	831	EPVEQDSSQPSLPLV	-0.28053	0.416727
		380	TPFIVPSSPTEQEGR	-0.57136	0.008651
TTK	Dual specificity protein kinase TTK	186	TAEKNASSPEKAKGR	0.140839	0.41381
		738	QPLTVVPVSPKFSTRF	1.107704	0.000479
TTK	Dual specificity protein kinase TTK	369	KICRDPQTPTVLQTKH	0.074426	0.84967
		19	AASAASGSPGPGEGS	-0.29259	0.514772
TTK	Dual specificity protein kinase TTK	697	TGVVAKLSPANQRKC	0.771591	0.088797
		473	SGVKRSRSGEGEVSG	-0.38443	0.018897
TTK	Dual specificity protein kinase TTK	436	VFSVSKQSPPISTSK	0.464797	0.018196

Appendix Table S1:

Table showing all proteins depicted in Fig. 6A, with the corresponding gene names, position of phosphosites detected, sequence window around the phosphosites and the results of enrichment in prolonged prophase after applying linear modeling for analysis.

Table EV1: Parameter values for the model of Rata et al. 2018, used for the simulations shown in Figure 1, 2 and EV2. Compared to the original work we rescaled all kinetic parameters, except those indicated in bold, by a factor of 0.025 to account for the duration of G2-phase of several hours (right column). The total cdk2-cyclin A concentration is varied to model the differences between cells.

Name	Name in code	Description	Value	Ratio to parameters in Rata et al. (2018)
k_{aPP1}	kapp1	Constitutive dephosphorylation and thereby activation of PP1	$0.01725 h^{-1}$	0.025
k_{aPP1a}	kapp1a	Dephosphorylation of PP1 by dephosphorylated PP1 in trans	$1.0581 h^{-1}$	0.025
k_{iPP1}	kipp1	Constitutive phosphorylation and hence inactivation of PP1	$0.0027 h^{-1}$	0.025
$k_{Cdk1,PP1}$	kipp1C	Phosphorylation of PP1 by Cdk1	$1.1323 h^{-1}$	0.025
$k_{PP1,Gwl}$	kPP1Gw	Dephosphorylation of Gwl by PP1	$27.7086 h^{-1}$	0.025
k_{ass}	kass	Association of pENSA with PP2A:B55	$925.9211 h^{-1}$	0.025
k_{dis}	kdis	Dissociation of the pENSA:PP2A:B55 complex	$0.0132 h^{-1}$	0.025
k_{catB55}	kcatB55	Dephosphorylation of pENSA when in complex with PP2A:B55	$1.5507 h^{-1}$	0.025
$k_{Gwl,ENSA}$	kGwENSA	Phosphorylation of ENSA by Gwl	$31.3216 h^{-1}$	0.025
$k_{PPX,Gwl}$	kppxGwl	Basal dephosphorylation of Gwl	$0.234 h^{-1}$	0.025
$k_{Cdk1,Sub}$	kcBc1Sub	Phosphorylation of the substrate by Cdk1	$0.12 h^{-1}$	0.250
$k_{B55,Sub}$	kB55Sub	Dephosphorylation of the substrate by PP2A:B55	$0.8895 h^{-1}$	0.250
$k_{Cdk1,Gwl}$	kcBc1G	Phosphorylation of Gwl by Cdk1	$0.35895 h^{-1}$	0.025
$k_{B55,Gwl}$	kB55G	Dephosphorylation of Gwl by PP2A:B55	$744.8454 h^{-1}$	0.025
$k_{Cdk2,Gwl}$	kcAc2G	Phosphorylation of Gwl by Cdk2	$0.2874 h^{-1}$	0.025
k_{Cdc25S}	k25S	Dephosphorylation of Y15 of Cdk1 by unphosphorylated Cdc25	$0.0075 h^{-1}$	0.025
k_{Cdc25F}	k25F	Dephosphorylation of Y15 of Cdk1 by phosphorylated Cdc25	$1.4117 h^{-1}$	0.025
k_{Wee1S}	kweeS	Phosphorylation of Y15 of Cdk1 by phosphorylated Wee1	$0.0075 h^{-1}$	0.025
k_{Wee1F}	kweeF	Phosphorylation of Y15 of Cdk1 by unphosphorylated Wee1	$70.9406 h^{-1}$	0.025
$k_{Cdk1,Wee1}$	kcBc1W1	Phosphorylation of Wee1 by Cdk1	$1.9698 h^{-1}$	0.025
$k_{Cdk1,Cdc25}$	kcBc125	Phosphorylation of Cdc25 by Cdk1	$0.19698 h^{-1}$	0.0025
$k_{PPX,Y15}$	kppxY15	Dephosphorylation of Wee1 and Cdc25 by a constitutive phosphatase	$0.0075 h^{-1}$	0.025
$k_{Cdk2,Wee1}$	kcAc2W1	Phosphorylation of Wee1 by Cdk2	$0.1644 h^{-1}$	0.025
$k_{Plk1,Cdc25}$	kplk1225	Phosphorylation of Cdc25 by Plk1	$0.01644 h^{-1}$	0.0025
$k_{B55,Wee1}$	kB55W1	Dephosphorylation of Wee1 by PP2A:B55	$0.82665 h^{-1}$	0.025
$k_{N55,Cdc25}$	kB5525	Dephosphorylation of Cdc25 by PP2A:B55	$0.82665 h^{-1}$	0.025
f_{Plk1}	fplk1	Fraction active Plk1	0 - 1	NA
$CycBCdk1_{Tot}$	CycBCdk1T	Total Cdk1 Cyclin B complexes	8.1808 a.u.	1
$CycACdk2_{Tot}$	CycACdk2T	Total Cdk2 Cyclin A complexes	2 - 5 a.u.	2-5
$PP1_{Tot}$	PP1T	Total PP1	1 a.u.	1
Gwl_{Tot}	Gwtot	Total Gwl	1 a.u.	1
$ENSA_{Tot}$	ENSAtot	Total Ensa	1 a.u.	1
$B55_{Tot}$	B55tot	Total B55	0.25 a.u.	1
Sub_{Tot}	SubT	Total Cdk1 phosphorylation substrate	1 a.u.	1