Supplemental material



Zhong et al., http://www.jcb.org/cgi/content/full/jcb.201208060/DC1

Figure S1. **Cdc7 function regulates replication fork progression.** (A) Experimental scheme: WT and cdc7-as3 cells were synchronized in G1 phase with α -factor for 4 h, treated with PP1 25 min before release, and released from α -factor in the presence of PP1 and 400 µg/ml BrdU. (B) Aliquots of the cultures were harvested for analysis by BrdU-IP-chip at the indicated times. (C) Experimental scheme: cdc7-as3 cells were synchronized in G1 phase with α -factor for 4 h, treated or not treated with PP1 25 min before release, and released from α -factor in the presence or absence of PP1 and presence of 0.033% MMS. (D) Samples were withdrawn at the indicated times for DNA content analysis by FACScan. (E) Aliquots of the cultures were pulsed with BrdU for the indicated intervals and harvested for analysis by BrdU-IP-chip. Data shown are from a single representative experiment out of two replicates, except the –PP1 sample in E, which was performed once.



Figure S2. Effective depletion of Cdc7 function with the cdc7-1 allele. (A) Experimental scheme: WT, cdc7-1, and cdc7-1 mcm5-bob1 cells were synchronized in G1 phase with α -factor for 3 h at 23°C, shifted to 32°C for 1 h, and released from α -factor at 32°C into the presence of 0.033% MMS. (B) Samples were withdrawn at the indicated times for DNA content analysis by FACScan. Data shown are from a single representative experiment out of two replicates.



Figure S3. **Deregulated origin firing in rad53** slows replication forks. (A) Experimental scheme: WT, rad53, exo1 Δ , and rad53 Δ exo1 Δ cells (all strains are sml1 Δ) were synchronized in G1 phase with α -factor for 4 h at 23°C and released from α -factor at 23°C into the presence of 0.033% MMS. (B) Samples were withdrawn at the indicated times for DNA content analysis by FACScan. (C) Aliquots of the cultures were pulsed with BrdU for the indicated intervals and harvested for analysis by BrdU-IP-chip. Data shown are from a single experiment.

Table S1.	Summary o	f data [.]	for f	fork I	rate	and	origin	firing	estimatio	n

Strain + condition	Experiment ID	Time point used to calculate number of origins firing	Time point used to calculate color for heat maps	Number of origins fired	Time points used for calculating fork rate	Fork rate
						bp/min
WT – MMS	cdc7as3-setA	1 + 2	1 + 2	233	1, 2, and 3	587
cdc7as3 - MMS	cdc7as3-setA	1 + 2	1 + 2	156	2 and 3	1,378
WT	cdc7as3-setA	1 + 2	1 + 2	223	1, 2, 3, and 4	478
cdc7as3	cdc7as3-setA	1 + 2	1 + 2	152	1, 2, 3, and 4	1,048
WT – MMS	cdc7as3-setB	1 + 2	1 + 2	234	1 and 2	199
cdc7as3 - MMS	cdc7as3-setB	1 + 2	1 + 2	157	1, 2, and 3	1,051
WT	cdc7as3-setB	1 + 2	1 + 2	214	1, 2, 3, and 4	414
cdc7as3	cdc7as3-setB	1 + 2	1 + 2	125	1, 2, and 3	1,014
WT	cdc7-1-setA	1	1 + 2	238	1, 2, 3, and 4	533
cdc7-1/mcm5- bob1	cdc7-1-setA	1	1 + 2	178	1 and 2	1,214
cdc7-1/mcm5- bob1 pph3	cdc7-1-setA	1	1 + 2	176	1, 2, and 3	973
WT	cdc7-1-setB	ND	ND	ND	1, 2, 3, and 4	549
cdc7-1/mcm5- bob1	cdc7-1-setB	ND	ND	ND	1 and 2	1,514
WT	cdc7-1-setC	1	1 + 2	253	ND	ND
cdc7-1/mcm5- bob1	cdc7-1-setC	1	1 + 2	179	ND	ND
cdc7-1/mcm5- bob1 pph3	cdc7-1-setC	1	1 + 2	161	ND	ND
WT	orc1-setA	1 + 2	1 + 2	224	1, 2, 3, and 4	580
orc1-161	orc1-setA	1 + 2	1 + 2	183	1 and 2	1,171
WT	orc1-setB	1 + 2	1 + 2	236	1, 2, 3, and 4	883
orc1-161	orc1-setB	1 + 2	1 + 2	201	1 and 2	1,232
WT	mec1-setA	1 + 2	1 + 2	205	2, 3, and 4	454
mecl	mec1-setA	1 + 2	1 + 2	301	2, 3, and 4	250
WT	mec1-setB	1 + 2	1 + 2	232	1, 2, 3, and 4	579
mecl	mec1-setB	1 + 2	1 + 2	318	1, 2, and 3	233