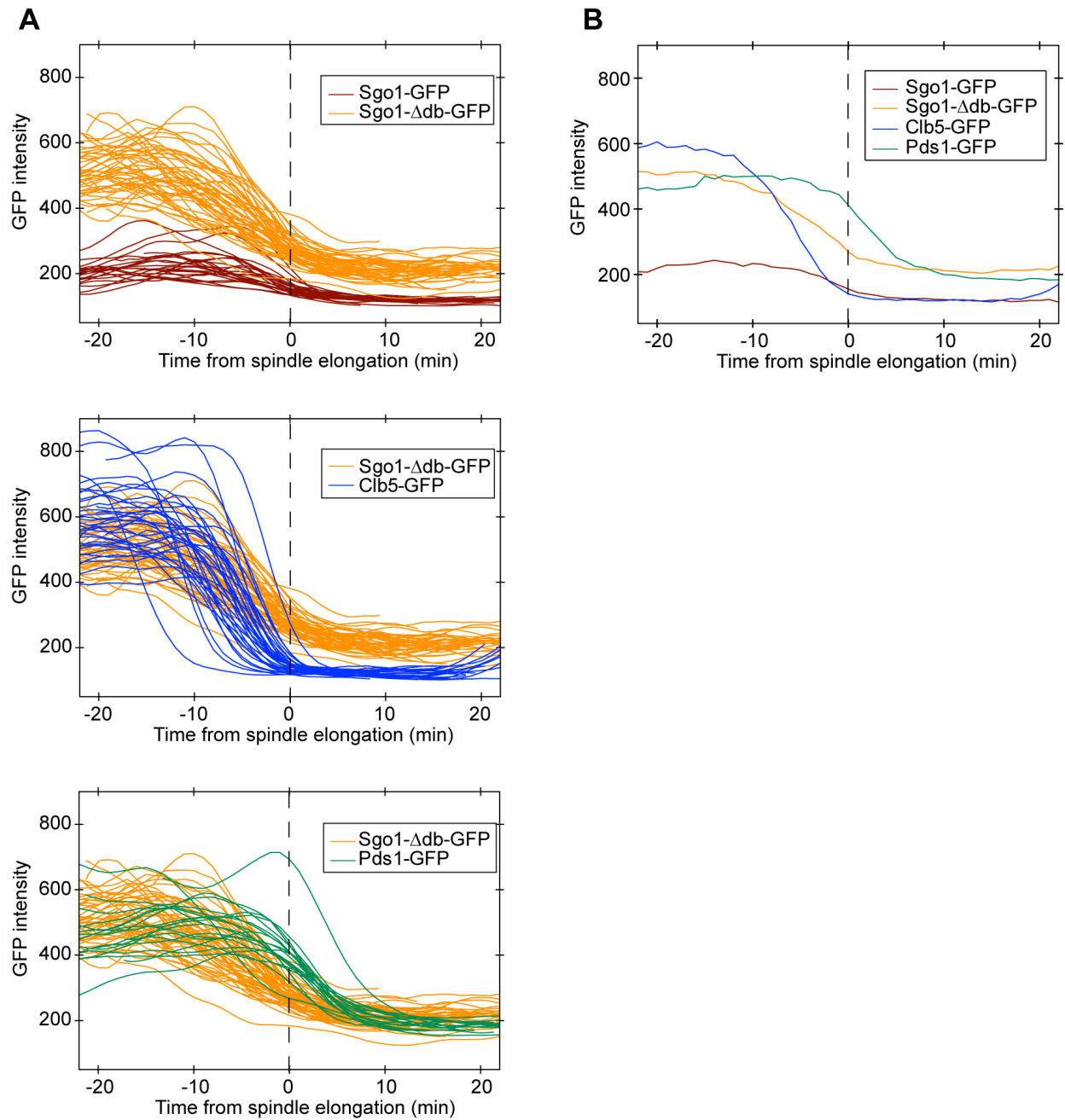**Figure S1. Sgo1 is a substrate of Cdc20 and Cdh1 *in vitro*.** Radiolabeled Sgo1 or Sgo1-Δdb was translated *in vitro* and tested for APC/C-dependent ubiquitination as in Fig. 1A, using either purified Cdc20 or Cdh1 as the activator subunit. All reactions contained E1 and E2. Cdh1 is generally more stable and active than Cdc20 in these assays, resulting in higher activity.

Figure S2. Un-normalized traces of GFP-labeled proteins. (A) Smoothed fluorescence intensity measurements (arbitrary units) of Sgo1-GFP, Sgo1Δdb-GFP, Clb5-GFP, or Pds1-GFP before normalization for the plots in Fig. 2. (B) Average intensity of un-normalized traces.

Table S1. Strains used in this study.

Strain	Genotype
HE153	*SGO1:URA3*
HE156	*SGO1- db:URA3*
HE167	*SGO1-GFP:KanMX4, SPC42-mCherry:HIS3*
HE168	*SGO1- db-GFP:KanMX4, SPC42-mCherry:HIS3*
DL009C	*CLB5-GFP:URA3, SPC42-mCherry:HIS3*
DL009P	*PDS1-GFP:URA3, SPC42-mCherry:HIS3*
HE297	*sgo1-3A- db-GFP:KanMX4, SPC42-mCherry:HIS3*
HE217	*SGO1- db:URA3, RTS1-GFP:KanMX4, SPC42-mCherry:HIS3*
HE213	*sgo1-3A- db:URA3, RTS1-GFP:KanMX4, SPC42-mCherry:HIS3*
HE294	*SGO1- db-GFP:KanMX4, SPC42-mCherry:HIS3, pGAL-CDC20:NatNT2*
HE237	*SGO1-9myc:HIS3*
HE239	*sgo1-3A-9myc:HIS3*
HE234	*RTS1-6HA:NatNT2*
HE241	*SGO1-9myc:HIS3, RTS1-6HA:NatNT2*
HE245	*sgo1-3A-9myc:HIS3, RTS1-6HA:NatNT2*
HE211	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI:HIS3*
HE212	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI-Rts1:HIS3*
HE207	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI:HIS3, sgo1 :NatNT2*
HE209	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI-Rts1:HIS3, sgo1 :NatNT2*
HE305	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI:cgHIS3*
HE307	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI:cgHIS3, sgo1 :NatNT2*
HE276	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI:cgHIS3, sgo1-3A:URA3*
HE266	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI:cgHIS3, rts1 :NatNT2*
HE309	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI-Rts1:cgHIS3*
HE311	*CEN5::256xlacO:LEU2, his3::pCUP1-GFP-LacI-Rts1:cgHIS3, sgo1 :NatNT2*
HE312	*CEN5::96xtetO:TRP1, leu2::pCUP1-GFP-TetR:cgLEU2, his3::pCUP1-GFP-LacI:cgHIS3*
HE314	*CEN5::96xtetO:TRP1, leu2::pCUP1-GFP-TetR:cgLEU2, his3::pCUP1-GFP-LacI:cgHIS3, sgo1 :NatNT2*
HE316	*CEN5::96xtetO:TRP1, leu2::pCUP1-GFP-TetR:cgLEU2, his3::pCUP1-GFP-LacI-Rts1:cgHIS3*
HE318	*CEN5::96xtetO:TRP1, leu2::pCUP1-GFP-TetR:cgLEU2, his3::pCUP1-GFP-LacI-Rts1:cgHIS3, sgo1 :NatNT2*