Derive et al., http://www.jcb.org/cgi/content/full/jcb.201504059/DC1

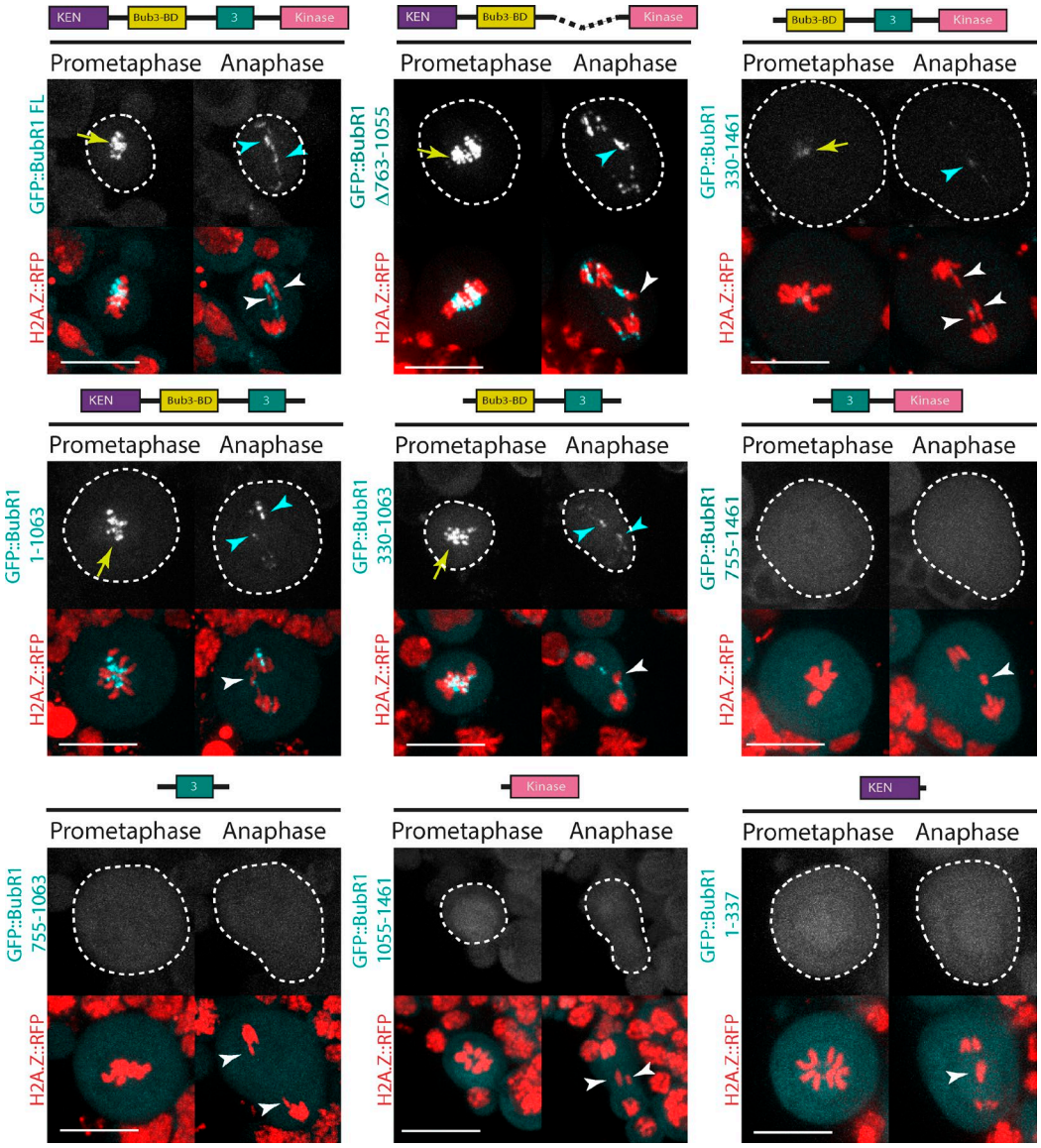


Figure S1. **The Bub3-BD of BubR1 is required for its localization on both the kinetochore and tether.** Time-lapse images of neuroblasts expressing H2A.Z::RFP and the indicated GFP::BubR1 truncated construct after I-CreI expression. The kinetochore and tether localizations of the GFP::BuBR1 constructs are indicated with yellow arrows and cyan arrowheads, respectively. The white arrowheads point to the I-CreI–induced acentric chromatids. The cells are delineated with dotted lines. Bars, 10 µm.

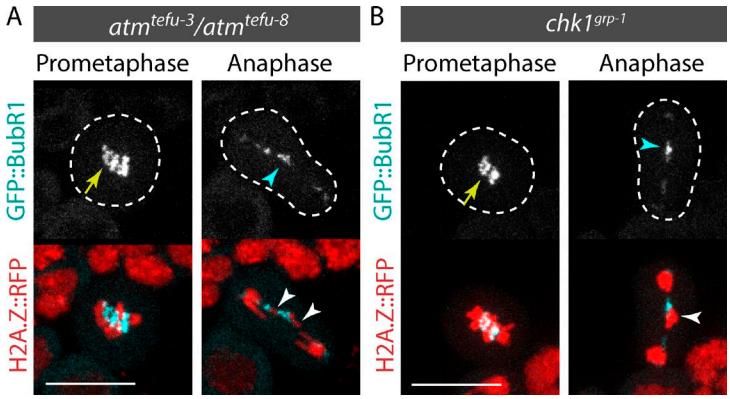


Figure S2. **The DNA damage checkpoint kinases Chk1 and ATM are not required for BubR1 recruitment to DNA breaks.** Time-lapse images of *atm*$^{tefu-3}$/*atm*$^{tefu-8}$ and *chk1*$^{grp-1}$ mutant neuroblasts expressing I-CreI and labeled with H2A.Z::RFP and GFP::BubR1. The yellow arrows indicate the localization of GFP::BubR1 on the kinetochore during prometaphase. The cyan arrowheads point to the GFP::BubR1 signal on the tether. The white arrowheads designate the broken chromatids. The cells are delineated with dotted lines. Bars, 10 µm.

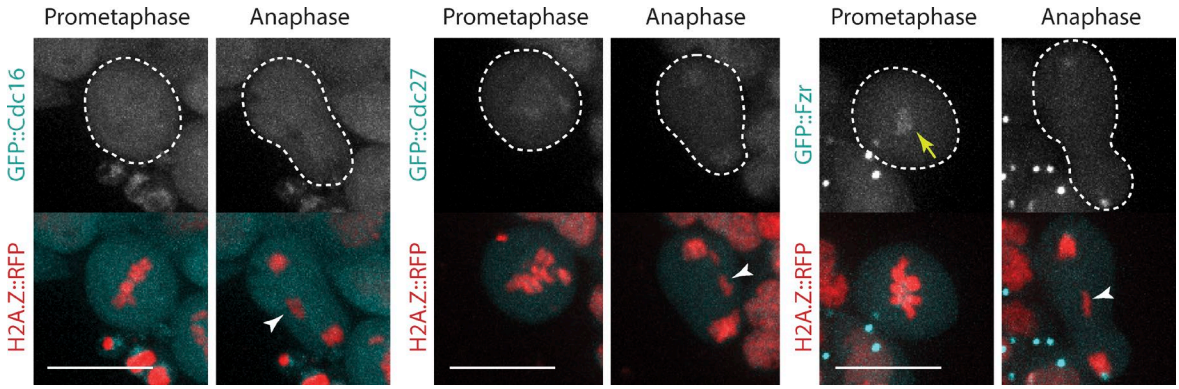


Figure S3. **The APC/C subunits Cdc16, Cdc27, and cofactor Fzr do not localize on DNA breaks.** Time-lapse images of neuroblasts expressing H2A.Z::RFP and GFP::Cdc16, GFP::Cdc27, and GFP::Fzr (left, middle, and right) after I-CreI expression. The white arrowheads point to the broken chromatids. The yellow arrow indicates the localization of GFP::Fzr on the kinetochore during prometaphase. The cells are delineated with dotted lines. Bars, 10 µm.

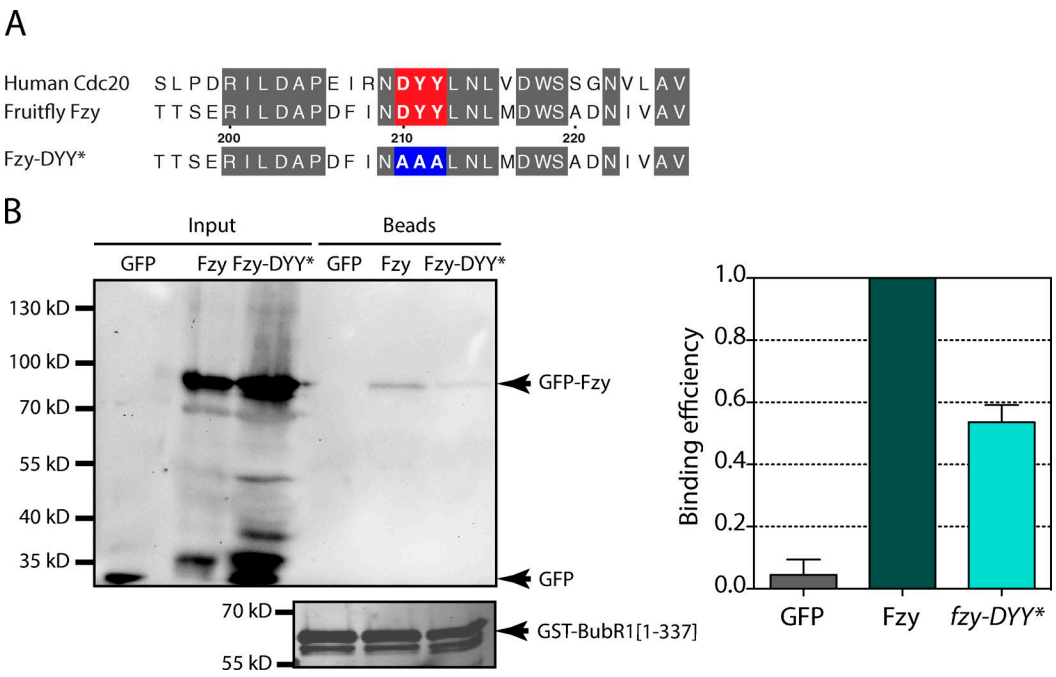


Figure S4. **The substitution of D210, Y211, and Y212 by alanine affects Fzy association with BubR1 N-terminal sequence.** (A) Sequence alignment of human Cdc20 and fruit fly Fzy around the DYY residues, which have been identified in human Cdc20 as essential for Cdc20 interaction with the first KEN box of human BubR1. These residues correspond to D210, Y211, and Y212 in *Drosophila* Fzy. We mutated these three residues into alanine and called the resulted mutant Fzy-DYY*. (B) Western blot showing the in vitro pull-down of GFP, GFP::Fzy (Fzy), and GFP::Fzy-DYY* (Fzy-DYY*) with GST::BubR1 [1–337]. The first three lanes show the level of GFP-labeled proteins extracted from embryos (Input). The last three lanes show the level of GFP-labeled proteins bound to the beads. The histogram corresponds to the binding efficiency of GFP and Fzy-DYY* to GST::BubR1 [1–337] relative to Fzy from three independent experiments.

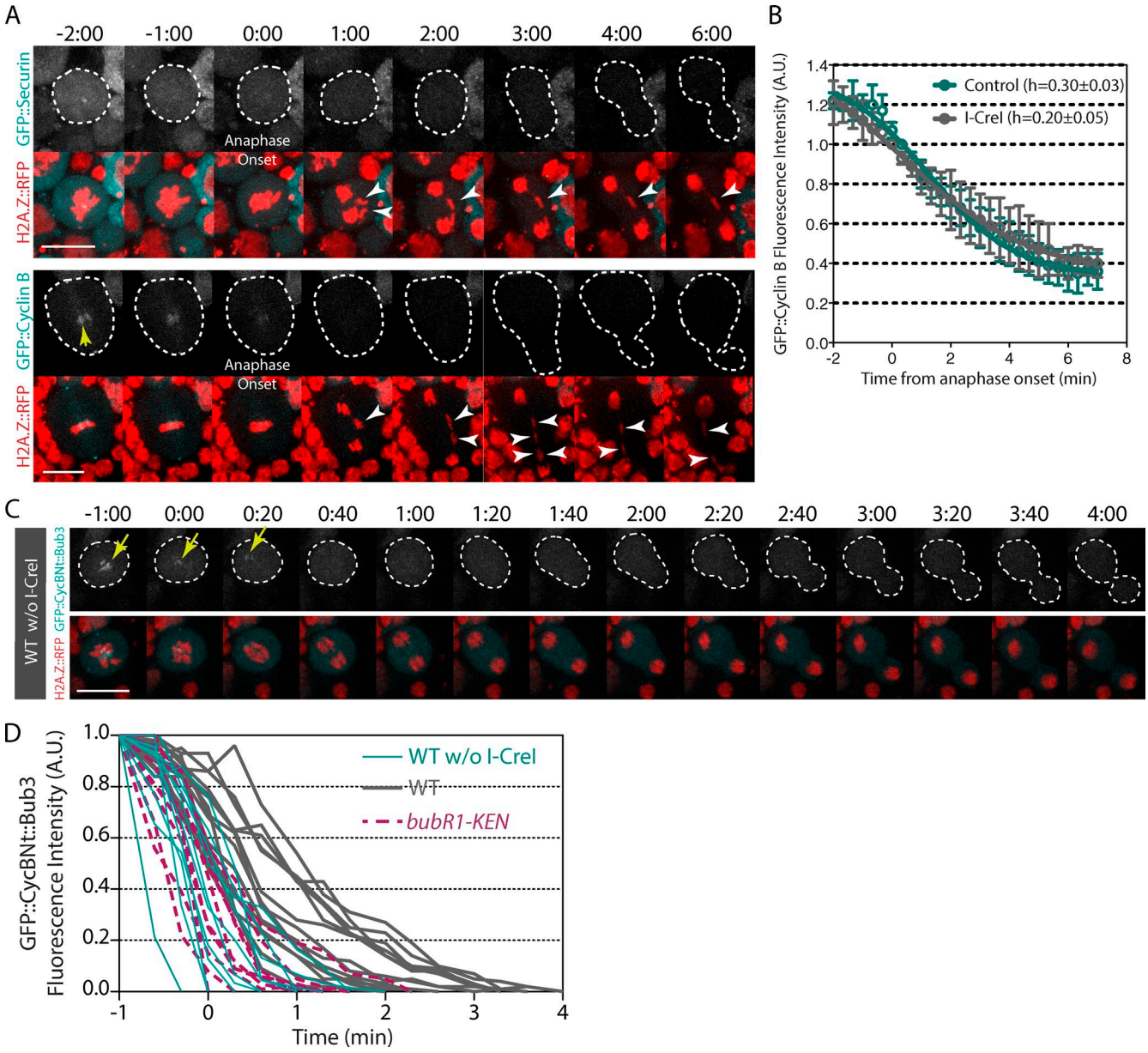


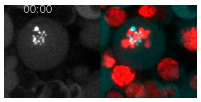
Figure S5. **The dynamics of GFP::Securin and GFP::Cyclin B during anaphase after I-CreI expression.** (A and B) Securin and Cyclin B do not localize on the broken chromatids throughout mitosis. (A) Time-lapse images of neuroblasts expressing I-CreI and labeled with H2A.Z::RFP and GFP::Securin (top) or GFP::Cyclin B (bottom). The kinetochore localization of GFP::Cyclin B is indicated with a yellow arrow. The white arrowheads point to the I-CreI–induced acentric chromatids. The cells are delineated with white dotted lines. Time (given in minutes/seconds) 0:00 corresponds to anaphase onset. Bars, 10 µm. (B) Quantitative analysis of the disappearance of the GFP::Cyclin B signal in control cells and in cells expressing I-CreI. The graph shows the average fluorescence intensities of GFP signals over time every 20 s. The fluorescence intensities were normalized with the fluorescence intensity measured at anaphase onset. (C and D) The APC/C synthetic substrate GFP::CycBNt::Bub3 is maintained on the tether during early anaphase in a BubR1 KEN box–dependent manner. (C) Time-lapse images of a WT cell expressing GFP::CycBNt::Bub3 without I-CreI induction (WT no HS). GFP::CycBNt::Bub3 localizes on the kinetochore during metaphase (yellow arrows). Upon anaphase onset, the GFP::CycBNt::Bub3 signal disappears from the kinetochore within 40 s. Time 0:00 corresponds to anaphase onset. (D) Quantitative analysis of the disappearance of the GFP::CycBNt::Bub3 signal on the kinetochore and tether for WT without I-CreI induction (WT no HS), WT, and *bubR1-KEN* mutant cells after I-CreI induction. The graph shows the fluorescence intensities of GFP signals on kinetochores and tethers over time for each cell. The fluorescence intensities were normalized with the fluorescence intensity measured at the time point −1:00 min. The GFP signal was measured over time every 20 s. The time starts at anaphase onset. This figure shows the raw data of Fig. 6 C for WT and *bubR1-KEN* mutant after I-CreI induction. Error bars represent mean ± 95% confidence interval. A.U., arbitrary units.

Table S1. **Genotypes of the larvae used for each figure**

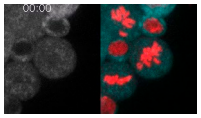
Genotype	Short name	Figure
$y^1w^{1,118}$; *bubR1^1/bubR1^1*; p[w$^+$, ubi>EGFP::BubR1 truncated]/ p[w$^+$, H2A.Z-mRFP], p[v$^+$, hsp70>I-CreI]	BubR1	1 and S1
$y^1w^{1,118}$; +; p[w$^+$, ubi>EGFP::BubR1 330-762[E481K]]/ p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]	330–762 [E481K]	1
$y^1w^{1,118}$;+; p[w$^+$, H2A.Z::EGFP], p[v$^+$, hsp70>I-CreI]/p[w$^+$, bub3>mRFP::Bub3]	RFP::Bub3 H2A.Z::GFP	2 A
$y^1w^{1,118}$; *bubR1^1/bubR1^1*; p[w$^+$, bub3>mRFP::Bub3]/ p[w$^+$, H2A.Z::EGFP], p[v$^+$, hsp70>I-CreI]	*bubR1^1*	2 B
$y^1w^{1,118}$, p[w$^+$, bubR1>EGFP::BubR1], p[v$^+$, hsp70>I-CreI]/+; p[w$^+$, H2A.Z::mRFP]/+ ; *bub3^1/ bub3^1*	*bub3^1*	2 C
$y^1w^{1,118}$; p[w$^+$, bub3>EGFP::Bub3]/+; p[w$^+$, H2A.Z::mRFP]/+	GFP::Bub3 H2A.Z::RFP	2 D
$y^1w^{1,118}$, p[w$^+$, bubR1>EGFP::BubR1]/ $y^1w^{1,118}$;+; p[w$^+$, H2A.Z::mRFP]/+	GFP::BubR1 H2A.Z::RFP	2 E
$y^1w^{1,118}$;+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/p[w$^+$, spc105> Spc105::EGFP]	Spc105::GFP H2A.Z::RFP	3 A
$y^1w^{1,118}$;+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/p[w$^+$, cenpC>CenpC::EGFP]	CenpC::GFP H2A.Z::RFP	3 B
$y^1w^{1,118}$;+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/p[w$^+$, g>EGFP::Nuf2]	GFP::Nuf2 H2A.Z::RFP	3 C
$y^1w^{1,118}$; p[w$^+$, mad1> Mad1::EGFP]/+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	Mad1::GFP H2A.Z::RFP	3 D
$y^1w^{1,118}$, p[w$^+$, bubR1>GFP::BubR1], p[v$^+$, hsp70>I-CreI]/ $y^1w^{1,118}$;+;p[w$^+$, H2A.Z::mRFP], *mad1/ mad1*	*mad1*	3 E
$y^1w^{1,118}$; p[w$^+$, mps1>EGFP::Mps1]/+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	GFP::Mps1 H2A.Z::RFP	3 F
$y^1w^{1,118}$; p[w+, ubi>RFP::BubR1]; *mps1^1*	*mps1^1*	3 G
$y^1w^{1,118}$; p[w$^+$, ubi>GFP::BubR1-KARD-D]/+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*BubR1-KARD-D*	3 H
$y^1w^{1,118}$; p[w$^+$, ubi>GFP::BubR1-KARD-A]/+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*BubR1-KARD-A*	3 I
$y^1w^{1,118}$; p[w$^+$, ubi>EGFP::Fzy]/+ ; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	WT	4, A–D
$y^1w^{1,118}$, p[w$^+$, bubR1>mRFP::BubR1-KEN]; *bubR1^1*, p[w+, ubi>EGFP::Fzy]*/bubR1^1*; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*bubR1-KEN*	4, A–D
$y^1w^{1,118}$; p[w$^+$, ubi>EGFP::Fzy-DYY*], *fzy^3*/+ ; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*fzy-DYY**	4, A–D
$y^1w^{1,118}$;+; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	WT	5
$y^1w^{1,118}$, p[v$^+$, hsp70>I-CreI]/ $y^1w^{1,118}$; p[w$^+$, H2A.Z::mRFP]/+ ; *bub3^1/ bub3^1*	*bub3^1*	5
$y^1w^{1,118}$, p[w+, bubR1>mRFP::BubR1-KEN]/ $y^1w^{1,118}$, p[w$^+$, bubR1>mRFP::BubR1-KEN]; *bubR1^1/ bubR1^1*; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*bubR1-KEN(2X)*	5
$y^1w^{1,118}$; *fzy^3*/+ ; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*fzy^3*/+	5
$y^1w^{1,118}$; p[w$^+$, ubi>EGFP::Fzy-DYY*], *fzy^3*/+ ; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*fzy^3*/+ Fzy-DYY*	5
$y^1w^{1,118}$; p[w$^+$, ubi>EGFP::Fzy-DYY*], *fzy^3*/ p[w$^+$, ubi>EGFP::Fzy-DYY*] ; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*fzy^3*/+ Fzy-DYY* (2X)	5
$y^1w^{1,118}$; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/ p[w$^+$, ubi>EGFP::CycBNt::Bub3]	WT	6
$y^1w^{1,118}$, p[w$^+$, bubR1>mRFP::BubR1::KEN]; *bubR1^1/bubR1^1*; p[w+, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/ p[w+, ubi>EGFP::CycBNt::Bub3]	*bubR1-KEN*	6
$y^1w^{1,118}$, p[w$^+$, bubR1>GFP::BubR1], p[v$^+$, hsp70>I-CreI]/+; p[w$^+$, H2A.Z::mRFP]/+; *tefu^{atm-3}/tefu^{atm-8}*	*atm^{tefu-3}/atm^{tefu-8}*	S2
$y^1w^{1,118}$, p[w$^+$, bubR1>GFP::BubR1]/+; *chk1^{grp-1}*; p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]/+	*chk1^{grp-1}*	S2
$y^1w^{1,118}$; p[w$^+$, ubi>GFP::Cdc16]/p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]	GFP::Cdc16 H2A.Z::RFP	S3
$y^1w^{1,118}$; +; p[w$^+$, ubi>GFP::Cdc27]/p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]	GFP::Cdc27 H2A.Z::RFP	S3
$y^1w^{1,118}$; +; p[w$^+$, ubi >GFP::Fzr]/p[w$^+$, H2A.Z::mRFP], p[v$^+$, hsp70>I-CreI]	GFP::Fzr H2A.Z::RFP	S3
$y^1w^{1,118}$; p[w$^+$, ubi >EGFP]/CyOy+	GFP	S4
$y^1w^{1,118}$; p[w+, ubi>EGFP::Fzy], *fzy^3*/CyOy+	Fzy	S4
$y^1w^{1,118}$; p[w+, ubi >EGFP::Fzy-DYY*], *fzy^3*/ CyOy+	*fzy-DYY**	S4
$y^1w^{1,118}$; p[w+, H2A.Z::mRFP], p[v+, hsp70>I-CreI]/ p[w+, Ub>EGFP::Securin]	GFP::Securin H2A.Z::RFP	S5 A
yw; p[w+, Ub>EGFP::Cyclin B]/+; p[w+, H2A.Z::mRFP], p[v+, hsp70>I-CreI]/ +	GFP::CyclinB H2A.Z::RFP	S5, A and B
yw; p[w+, Ub>EGFP::Cyclin B]/+; p[w+, H2A.Z::mRFP], p[v+, hsp70>I-CreI]/ + no heat shocked and yw; p[w+, Ub>EGFP::Cyclin B]/+; p[w+, H2A.Z::mRFP] heat shocked	Control	S5 B
yw; p[w+, Ub>EGFP::Cyclin B]/+; p[w+, H2A.Z::mRFP], p[v+, hsp70>I-CreI]/ +	I-CreI	S5 B
yw; p[w+, H2A.Z::mRFP], p[v+, hsp70>I-CreI]/ p[w+, Ub>EGFP::CycBNt::Bub3]	WT No HS	S5, C and D

Table S2. **Oligonucleotide sequences used in this study**

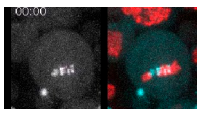
Oligonucleotide	Sequence (5′ to 3′)	Construct
F1	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGACTTTGACAATGCGAAAGAG	BubR1 FL [1–1,063] [1–337] [Δ763–1,055] BubR1-KARD-D BubR1-KARD-A
F2	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGACGAACAAGCAATGCCACCTATT	[330–1,063] [330–762]
F3	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCCCGGGTGGTGACAACTTTTTGCGC	[755–1,461] [755–1,063]
F4	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCATAAAAAATTCCACACTGCTGATC	[1,055–1,461]
R1	GGGGACCACTTTGTACAAGAAAGCTGGGTCCTATTTCTGCAATATCGTGTTAAACTT	BubR1 FL [755–1,461] [1,055–1,461] [Δ763–1,055] BubR1-KARD-D BubR1-KARD-A
R2	GGGGACCACTTTGTACAAGAAAGCTGGGTCGATCAGCAGTGTGGAATTTTTTAT	[1–1,063] [330–1,063] [755–1,063]
R3	GGGGACCACTTTGTACAAGAAAGCTGGGTCCTAAATAGGTGGCATTGCTTGTTCGTC	[1–337]
R4	GGGGACCACTTTGTACAAGAAAGCTGGGTCGCGCAAAAAGTTGTCACCACCCGG	[330–762]
Δ3NtR	AGGGATCAGCAGTGTGGAATTTTTTATGCGCAAAAAGTTGTCACCACCCGG	[Δ763–1,055]
Δ3CTF	GAAAAACCGGGTGGTGACAACTTTTTGCGCATAAAAAATTCCACACTGCTGATC	[Δ763–1,055]
CycBNt-Gat-F	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGACGAACAAGCAATGCCACCTATTGGCGCCGGAGCTGGCGCC GGAGCGATGGCCGCCCTTGAAAAGAAC	Ub>EGFP-CycBNt-Bub3
Bub3-Lnk-CycBNt-R	TGGGGGACGCGCTCCGGCGCCAGCTCCGGCGCCGCCCAGTGGCTGTTTCTTCCA	Ub>EGFP-CycBNt-Bub3
Lnk-Bub3-F	GGCGCCGGAGCTGGCGCCGGAGCGCGTCCCCCAGAGTTCAAGCTTA	Ub>EGFP-CycBNt-Bub3
Bub3-R	GGGGACCACTTTGTACAAGAAAGCTGGGTCCTATTATTTCTGTTTAGTTTCCTGGTC	Ub>EGFP-CycBNt-Bub3
Bub3prom-F	ACTGGGTACCGATCTTCTTGACGGTGCTCACCTGGAG	g>mRFP-Bub3
Bub3prom-R	ACTGCTCGAGTTTGTCAAGTTTTCTGCTAGCAACTTG	g>mRFP-Bub3
Bub3gen-F	AGCTCCCGGGATGCGTCCCCCAGAGTTCAAGCTTAAC	g>mRFP-Bub3
Bub3gen-R	CTATTCTAGAAAAAGCCCAGGTTTATTATGGCCAACG	g>mRFP-Bub3
mRFP-F	ACTGCTCGAGATGGCCTCCTCCGAGGACGTC	g>mRFP-Bub3
mRFP-R	ACTGCCCGGGCGCTCCGGCGCCAGCTCCGGCGCCCCTAGGGGCGCCGGTGGAGTGGCGGCCCTCG	g>mRFP-Bub3
KARD-R(D)	GGTACCGCTCGTCGCCAAGCTTTGATCGCCGTGCTCATCGGTTTCCAATATGGTATCTAGTTGCTTTCGCAAA GTTGGAGA	BubR1-KARD-D
KARD-R(A)	GGTACCGCTCGTCGCCAAGCTTTGTGCGCCGTGCTCAGCGGTTTCCAATATGGTAGCTAGTTGCTTTCGCAAA GTTGGAGA	BubR1-KARD-A
KARD-F(D)	TCTCCAACTTTGCGAAAGCAACTAGATACCATATTGGAAACCGATGAGCACGGCGATCAAAGCTTGGCGACGA GCGGTACC	BubR1-KARD-D
KARD-F(A)	TCTCCAACTTTGCGAAAGCAACTAGCTACCATATTGGAAACCGCTGAGCACGGCGCACAAAGCTTGGCGACGA GCGGTACC	BubR1-KARD-A
Securin-Nt-F	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCGATCAGATTTTAAACAAGGAAA	Ub>EGFP-Securin
Securin-Nt-R	GGGGACCACTTTGTACAAGAAAGCTGGGTCCTAAAATAGAACATCAATGCCTTCCAA	Ub>EGFP-Securin
Securin-Ct-F	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCATGGATCAGATTTTAAACAAGGAAA	Ub>Securin-EGFP
Securin-Ct-R	GGGGACCACTTTGTACAAGAAAGCTGGGTCAAATAGAACATCAATGCCTTCCAA	Ub>Securin-EGFP



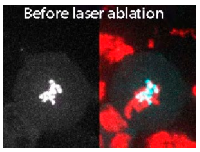
Video 1. **The BubR1-truncated construct 330–762 containing the Bub3-BD and fused with GFP localizes on I-CreI–induced DNA breaks during mitosis.** WT neuroblast expressing GFP::BubR1 330–762 (left, cyan on right) and H2A.Z::RFP (red) after I-CreI induction. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 1 C (left).



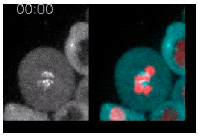
Video 2. **The BubR1 truncated construct 330–762 mutated on E481 does not localize on DNA breaks during mitosis.** WT neuroblast expressing GFP::BubR1 330–762 [E481K] (left, cyan on right) and H2A.Z::RFP (red) after I-CreI induction. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 1 C (right).



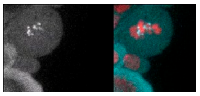
Video 3. **Bub3 localizes on I-CreI–induced DNA breaks.** WT neuroblast expressing RFP::Bub3 (left, cyan on right) and H2A.Z::GFP (red) after I-CreI induction. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 2 A (left).



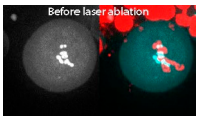
Video 4. **BubR1 localizes on laser-induced DNA breaks.** WT neuroblast expressing GFP::BubR1 (left, cyan on right) and H2A.Z::RFP (red). Image acquisition starts 20s before laser ablation, and the acquisition is paused while inducing chromosome breaks with a 355-nm pulsed laser and starts again. The arrows indicate the appearance of GFP::Bub3 signal on the breaks. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 2 E.



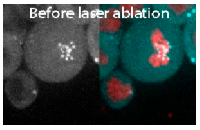
Video 5. **Fzy localizes on I-CreI–induced DNA breaks.** WT neuroblast expressing GFP::Fzy (left, cyan on right) and H2A.Z::RFP (red) after I-CreI induction. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 4 A (left).



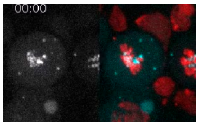
Video 6. **Fzy levels are reduced on I-CreI–induced DNA breaks in *bubR1-KEN* mutant.** *bubR1-KEN* mutant neuroblast expressing GFP::Fzy (left, cyan on right) and H2A.Z::RFP (red) after I-CreI induction. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 4 A (middle).



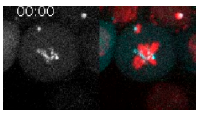
Video 7. **Fzy localizes on laser-induced DNA breaks.** WT neuroblast expressing GFP::Fzy (left, cyan on right) and H2A.Z::RFP (red) before and after laser-induced DNA breaks. The yellow circle indicates the site of laser ablation. The arrows indicate the appearance of GFP::Fzy signal on the breaks. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 4 B (top).



Video 8. **Fzy levels are reduced on laser-induced DNA breaks in *bubR1-KEN* mutant.** *bubR1-KEN* mutant neuroblast expressing GFP::Fzy (left, cyan on right) and H2A.Z::RFP (red) before and after laser-induced DNA breaks. The yellow circle indicates the site of laser ablation. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 4 B (middle).



Video 9. **The degradation of the synthetic APC/C substrate CycBNt::Bub3 is delayed on DNA breaks.** WT neuroblast expressing GFP::CycBNt::Bub3 (left, cyan on right) and H2A.Z::RFP (red) after I-CreI expression. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 6 B (top).



Video 10. **The degradation of the synthetic APC/C substrate CycBNt::Bub3 is not delayed on DNA breaks in *bubR1-KEN* mutant.** *bubR1-KEN* mutant neuroblast expressing GFP::CycBNt::Bub3 (left, cyan on right) and H2A.Z::RFP (red) after I-CreI expression. Images were analyzed by a time-lapse confocal spinning disk microscope equipped with an Evolve emCCD camera. Frames were taken every 20 s. Time is given in minutes/seconds. The video corresponds to Fig. 6 B (bottom).