Supplementary Figures and Movies

Fig S1: Dispersed chromatin and non-disjunction observed in cdh1 knockdown oocytes following maturation. (**a**, **b** and **c**) Chromatin (Hoechst, blue) and microtubule (anti-tubulin; red) staining in non-injected (a), *cdh1*^{MO} injected (b); and *cdh1*^{MO} + *cdh1* cRNA injected (c) oocytes. All 3 oocytes contained chromatin in a PB but only the non-injected and rescue (a and c) oocytes had chromatin aligned on a metaphase II spindle. Scale bar, 15μm. (**d, e**) Chromosome spreads for in vitro matured oocytes that were either non-injected (d) or injected with *cdh1*^{MO} (e). The non-injected spread shows 20 monovalent chromosomes, whereas the cdh1 knockdown oocyte is hyperploid with 22 monovalents, consistent with MI non-disjunction. Chromosome numbering relates to sequence ordering not to a karyotypic nomenclature.

Fig S2: Cdc20-dependent securin degradation in oocytes. **a,** Securin-CFP and cyclin B1-YFP fluorescence recorded in an oocyte during maturation. Note the simultaneous loss in securin and cyclin B1. **b,** Securin-GFP fluorescence during in vitro maturation of oocytes injected at the GV stage with or without *cdh1*^{MO} as indicated. **c,** calculated from (b), the timing of initiation of securin-GFP degradation after GVB for oocytes knocked down for cdh1 (*cdh1*^{MO}) or uninjected oocytes as indicated. Initiation of securin degradation is significantly earlier, marked by the asterisks, in *cdh1*^{MO} injected oocytes (t-test, p=0.001). **d**, Securin-GFP and cyclin B1-GFP degradation rates (arbitrary units per h) in control oocytes or those that have been microinjected with *cdh1*^{MO} as indicated. Securin and cyclin B1 degradation are unaffected by cdh1 knockdown (t-test, p=0.70 for securin and p=0.73 for cyclin B1). **e,** Securin-GFP degradation rates in *cdc20*^{MO} injected oocytes

(n=15) compared to non-injected (n=10). Cdc20 knockdown reduced the rate of securin B1 degradation, normalised with respect to the maximum fluorescence achieved during maturation (100%). Asterisks for each timepoint indicates the securin-GFP fluorescence in the *cdc20*^{MO} injected oocyte is significantly different from the uninjected ($p<0.05$, t-test).

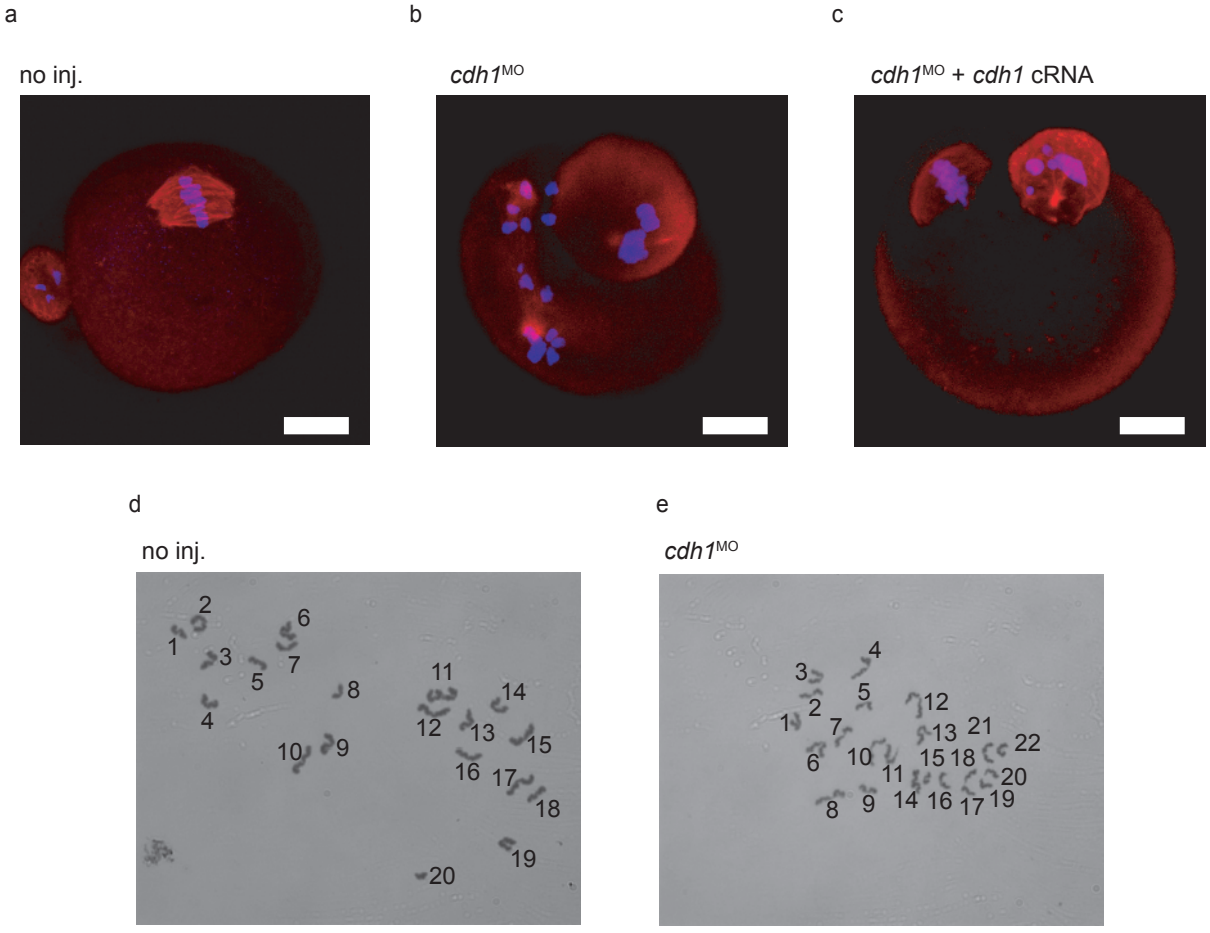
Fig S3: Cdc20-fluorescent protein degradation during oocyte maturation. **a**, Cdc20-GFP fluorescence levels during maturation were normalised with respect to the peak level achieved (F_{max}) before any cdc20 degradation, and the minimum fluorescence during oocyte maturation determined (F_{min}). **b**, Observed percentage loss in cdc20-GFP during maturation in control, cdh1 knockdown and cdh1 rescue oocytes. If no observed decrease was observed during maturation, the cdc20-GFP loss was set to zero. **c**, The timing of initiation of cyclin B1-YFP and cdc20-CeFP degradation, relative to GVB, with or without *cdh1*^{MO} microinjection. Loss of cdh1 brought forward cyclin B1 degradation to the time at which cdc20 was normally degraded. Error bars are s.d., and number of oocytes in parenthesis, pooled from 2 independent experiments per condition. **d**, Regression plot of onset of cyclin B1-YFP degradation against cdc20-CeFP degradation, relative to GVB, in oocytes expressing both constructs. The plot demonstrates a good correlation between the onset timings of their degradation ($r=0.842$, $p<0.001$). Regardless of the actual time of cdc20 degradation, cyclin B1 degradation started about 2 h later.

Fig S4: Full scans of key Western blots. *non-specific bands detected by some batches of anti-cdc20 antibody.

Supplementary Video 1: Maturation in a control non-injected oocyte that has been incubated in Hoechst 33258 to stain chromatin. The top panel is brightfield and the bottom panel is Hoechst fluorescence. GVB occurs at 1 h, cortical movement of the chromatin at 9.5 h, and PB extrusion at 10.25 h. Note the long period in which chromatin appears to be congressed on a metaphase plate in the centre of the oocyte, e.g. at 8.5 h. At the end of the movie the oocyte has arrested at metaphase II. Five frames per second; time bar is h: min (AVI; 2.9 MB; Quicktime compatible).

Supplementary Video 2: Maturation in an oocyte that has been microinjected with *cdh1*^{MO} at the GV stage and incubated in Hoechst 33258 to stain chromatin. The top panel is brightfield and the bottom panel in Hoechst fluorescence. GVB occurs at 0.75 h, cortical movement of the chromatin at 8.5 h, and PB extrusion at 9 h. Note that no alignment of chromatin happens before PB extrusion. Instead non-aligned chromatin moves to the oocyte cortex and is mis-segregated, with more chromatin being extruded into the PB than remaining in the oocytes (see 10.25 h). Lagging chromosomes are observed at 9.5 h onwards. Five frames per second; time bar is h: min (AVI; 2.5 MB; Quicktime compatible).

Fig. S1
Reis A


a
no inj.
b
cdh1MO
c
cdh1MO + cdh1 cRNA
d
no inj.
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
e
cdh1MO
1
2
3
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22

Fig. S2
Reis A

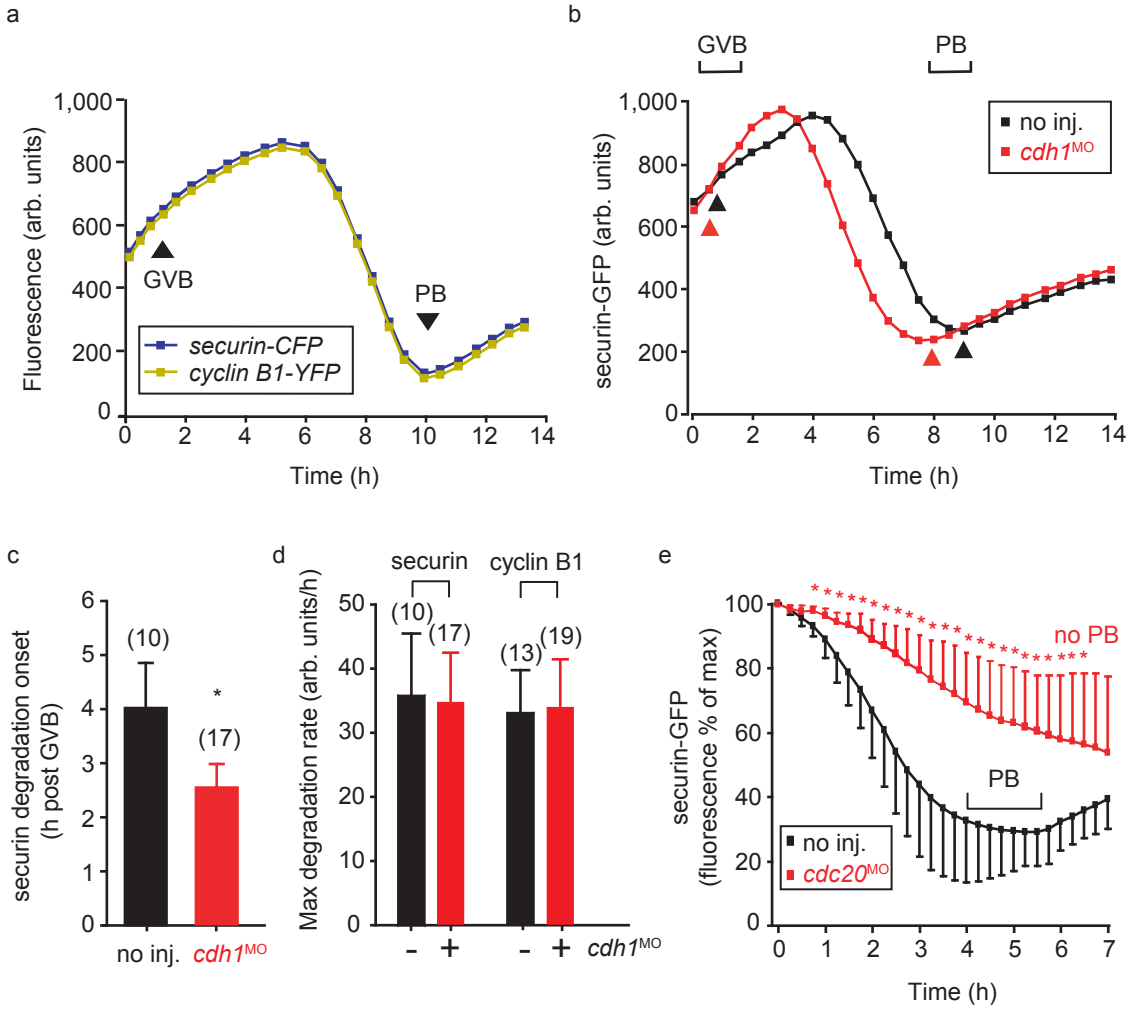
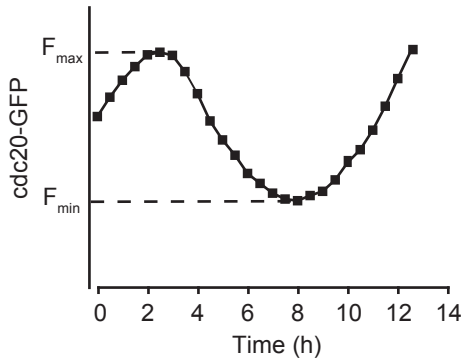

a
Fluorescence (arb. units)
1,000
800
600
400
200
0
GVB
PB
securin-CFP
cyclin B1-YFP
0 2 4 6 8 10 12 14
Time (h)
b
GVB
PB
securin-GFP (arb. units)
no inj.
cdh1MO
Time (h)
c
securin degradation onset
(h post GVB)
(10)
(17)
*
no inj.
cdh1MO
d
securin
cyclin B1
Max degradation rate (arb. units/h)
(10)
(17)
(13)
(19)
- + - +
cdh1MO
e
securin-GFP
(fluorescence % of max)
no PB
PB
no inj.
cdc20MO
Time (h)

Fig. S3
Reis A

a

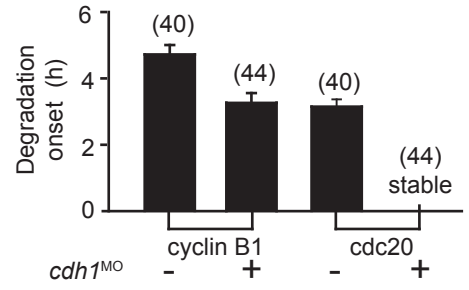


b

	Percentage cdc20-GFP degradation (F_{max}-F_{min}/F_{max})	n
control	30 +/- 8	22
cdh1^{MO}	0^{A}	16
cdh1^{MO} +*cdh1* cRNA	37 +/- 12	14

A, no degradation observed, so no F_{min}

c



d

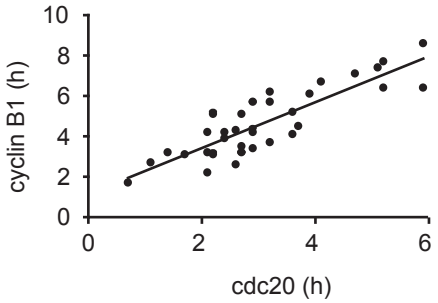
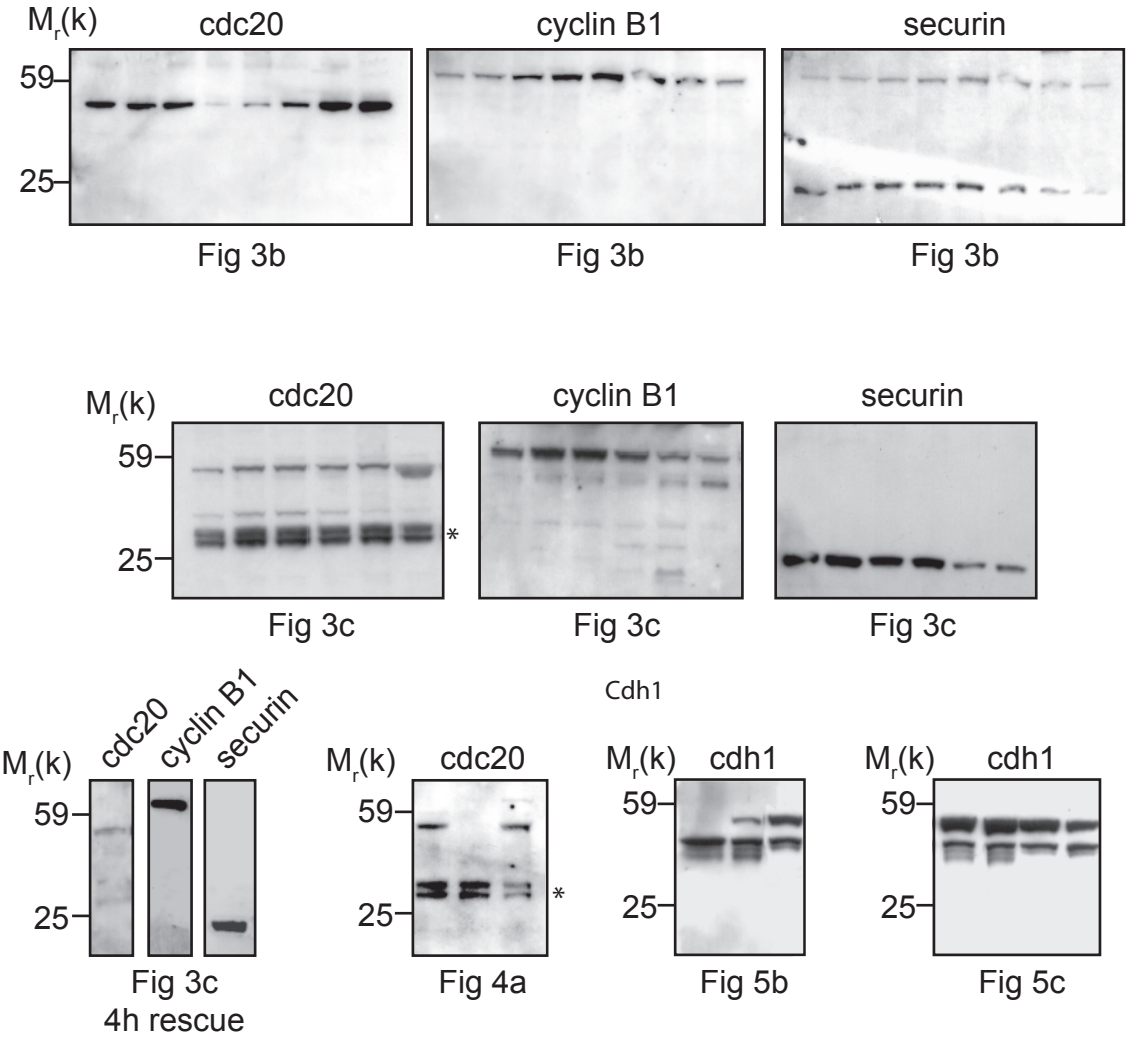


Fig S4


M$_r$(k)
cdc20
cyclin B1
securin
59
25
*
Fig 3b
Fig 3b
Fig 3b
Fig 3c
Fig 3c
Fig 3c
Fig 3c
4h rescue
Fig 4a
Cdh1
Fig 5b
Fig 5c