## Supplemental material

Ladouceur et al., http://www.jcb.org/cgi/content/full/jcb.201502092/DC1

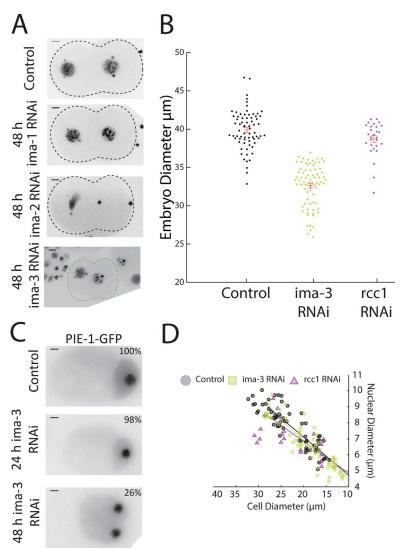
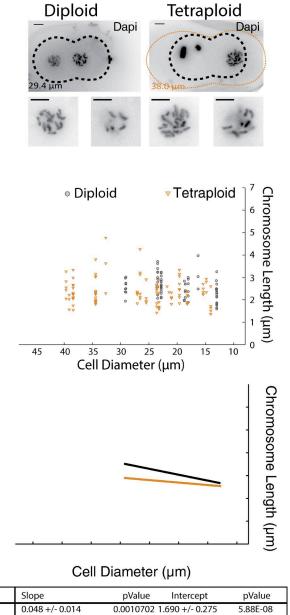


Figure S1. Embryo size differences for IMA-1 to -3 RNAi and RCC1 RNAi, PIE-1::GFP localization at the 4-cell stage, and nuclear to cell diameter relation in control, IMA-3, and RCC1 RNAi embryos. (A) Embryos depleted of the three importin- $\alpha$  proteins found in *C. elegans*. The outlines of the embryos are depicted with dashed black lines. (B) Embryo diameter in control, ima-3, or rcc1 RNAi. Error bars are standard error of the mean. (C) Percentage of embryos with the indicated localization or mislocalization of Pie-1::GFP in the P2 cell after 48- or 24-h depletion of ima-3. (A and C) Bars, 5 µm. (D) Correlation of cell to nucleus diameter in control or rcc1 RNAi embryos. Linear regression values are presented in Table S3.



Diploid 0.048 +/- 0.014 0.0010702 1.690 +/- 0.275 5.88E-08
Tetraploid 0.020 +/- 0.016 0.2044 1.892 +/- 0.342 6.34E-07

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Figure S2. **Chromosome length analysis in diploid versus tetraploid worms.** Representative still images of diploid and tetraploid embryos at the 2-cell stage. Shown is the outline of the embryo and size of the depicted embryo: black, diploid; orange, tetraploid. The diploid outline is overlaid on the tetraploid to illustrate differences in size. Bars, 5  $\mu$ m. Chromosome length correlated to cell size in diploid and tetraploid embryos from the 1- to 16-cell stage. Graph and table representing the linear regression of cell size versus chromosome length are shown. Diploid, n = 97; tetraploid, n = 156.

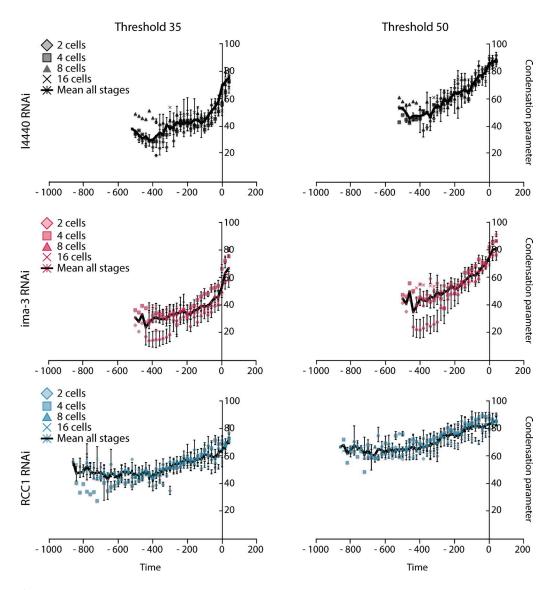


Figure S3. Plot of condensation dynamics at all developmental stages examined in this study. The number of pixels with a value below a threshold of either 35 or 50% is shown as indicated. Error bars are standard error of the mean.

Table S1.	One-way ANOVA	comparina devel	opmental stage for	r each RNAi condition	or all RNAi condition	s at each developmental stage

Condition/developmental stage	ANOVA significance
Condition	
Control RNAi	P < 0.0001
ima-3 RNAi	P < 0.0001
rcc1 RNAi	P < 0.0001
Developmental stage	
1-cell stage	P < 0.0001
2-cell stage	P < 0.0001
4-cell stage	P < 0.0001
8-cell stage	P < 0.0001
16-cell stage	P < 0.0001

One-way ANOVA comparing chromosome length differences measured in control, ima-3, and rcc1 RNAi at all developmental stages. The top half of the table shows the ANOVA significance for comparisons of chromosome length differences between all developmental stages from 1- to 16-cell stage for each RNAi condition. The bottom half of the table shows the ANOVA significance for comparisons of chromosome length differences between all RNAi conditions for each developmental stage from 1- to 16-cell stage. Using the Bonferroni correction gives a significance threshold of 0.01. Control RNAi, n = 354 chromosomes; IMA-3 RNAi, n = 287 chromosomes; RCC1 RNAi, n = 207 chromosomes.

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Table S2.	Student's t test statistica	l significance of chromos	ome lenath differences	comparing all devel	opmental stages for each RNAi condition

Developmental stage	Control RNAi	ima-3 RNAi	rcc1 RNAi
1 cell vs. 2 cell	ns	ns	P ≤ 0.05
1 cell vs. 4 cell	ns	$P \le 0.05$	$P \le 0.05$
1 cell vs. 8 cell	$P \le 0.05$	$P \le 0.05$	$P \le 0.05$
1 cell vs. 16 cell	$P \le 0.05$	$P \le 0.05$	$P \le 0.05$
2 cell vs. 4 cell	ns	ns	ns
2 cell vs. 16 cell	$P \le 0.05$	$P \le 0.05$	$P \le 0.05$
2 cell vs. 16 cell	$P \le 0.05$	$P \le 0.05$	ns
4 cell vs. 8 cell	$P \le 0.05$	ns	ns
4 cell vs. 16 cell	$P \le 0.05$	$P \le 0.05$	ns
8 cell vs. 16 cell	ns	$P \le 0.05$	ns

The multiple comparisons Student's t test was corrected using the Bonferroni correction. ns, not significant.

## Table S3. Correlation between cell size, nuclei size, and chromosome length in control and IMA-3-, RCC1-, and NTF-2-depleted embryos

Parameter	Slope	P-value	Intercept	
Call diameter vs. chromosome length (µm)				
Control (333)	0.052 ± 0.01	4.11 × 10 <sup>-7</sup>	2.056 ± 0.251	
ima-3 RNAi (285)	0.053 ± 0.007	1.38 × 10 <sup>-11</sup>	1.79 ± 0.144	
rcc1 RNAi (185)	0.019 ± 0.012	0.1016	1.857 ± 0.245	
Nuclear diameter vs. chromosome length (µm)				
Control (316)	0.130 ± 0.033	0.00011151	2.182 ± 0.252	
ima-3 RNAi (272)	0.210 ± 0.026	$4.28 \times 10^{-14}$	1.387 ± 0.179	
rcc1 RNAi (165)	0.120 ± 0.036	0.0010262	1.436 ± 0.264	
Cell diameter vs. nuclear diameter (µm)				
Control (64)	0.239 ± 0.022	$2.62 \times 10^{-15}$	2.334 ± 0.477	
ima-3 RNAi (70)	0.249 ± 0.016	7.13 × 10 <sup>-24</sup>	1.965 ± 0.291	
rcc1 RNAi (21)	0.199 ± 0.046	0.00056384	2.861 ± 0.954	
Cell volume vs. nuclear volume (µm <sup>3</sup> )				
Control (37)	0.009688 ± 0.0009577	$6.25 \times 10^{-12}$	183.55 ± 13.85	
rcc1 RNAi (23)	0.0053036 ± 0.0020787	0.0186	105.63 ± 27.965	
ntf-2 RNAi (27)	0.002163 ± 0.001139	0.0693	120.4 ± 13.22	

Bold indicates statistically different from control. P-value is the possibility of the slope being different than 0.

## Table S4. Time spent in prophase in control and RCC1-depleted embryos

RNAi	Mean time
4440	4 min 30 s
Rcc1	4 min 35 s

Time represents time of centrosome growth from duplication to NEBD.

Table S5	Statistical differences of chromosome length variance at all different stages between control and RCC1-depleted embryos
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Developmental stage	Variance control	Variance rcc1 RNAi embryos	Statistically different
	μm	μm	
1-cell stage	1.914	1.925	No
2-cell stage	1.751	2.113	No
4-cell stage	1.405	1.966	Yes
8-cell stage	1.858	1.226	No
16-cell stage	1.626	1.245	No