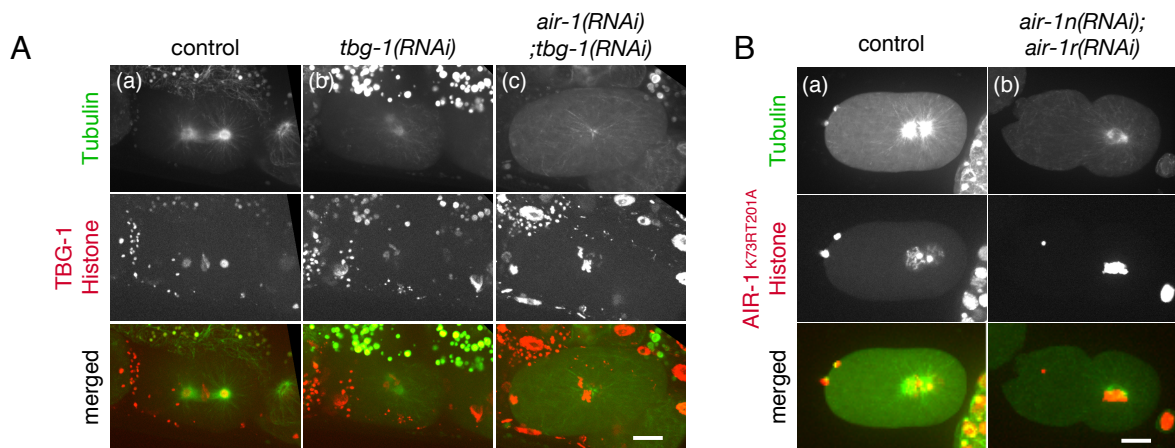


# Supplemental Materials

*Molecular Biology of the Cell*

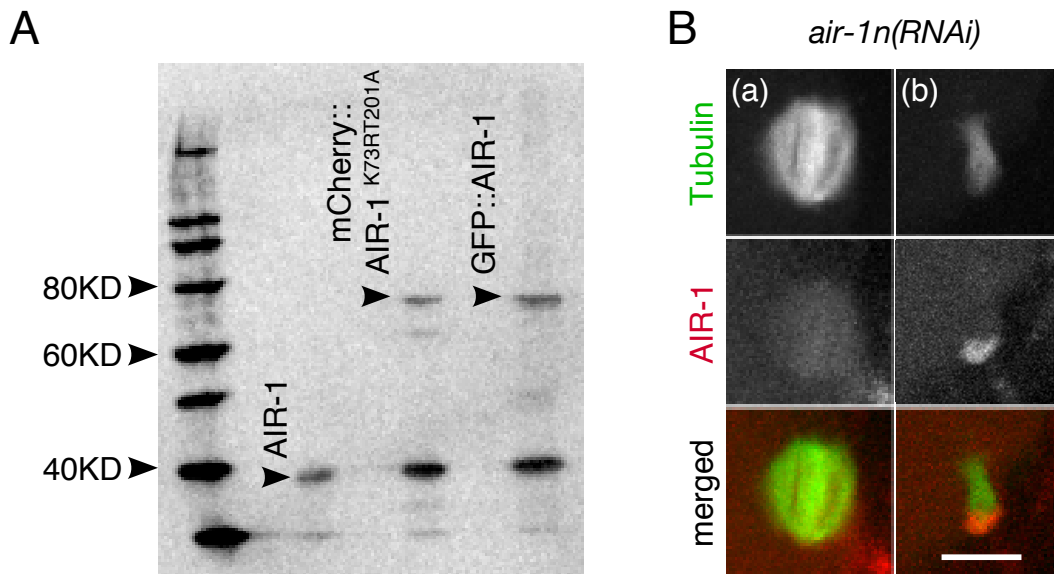
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## Supplementary Figures

**Figure S1. Confirmation of RNAi efficiency for *tbg-1* and *air-1***

A. Live images of 1-cell mitotic stage embryos expressing GFP:: $\beta$ -tubulin (green), mCherry::Histone (red), and mCherry::TBG-1 (red). (a) Control, (b) *tbg-1(RNAi)*, and (c) *air-1(RNAi);tbg-1(RNAi)*. Bar=10 $\mu$ m. In (b) and (c), the mCherry::TBG-1 signal is undetectable and monopolar spindles are formed.

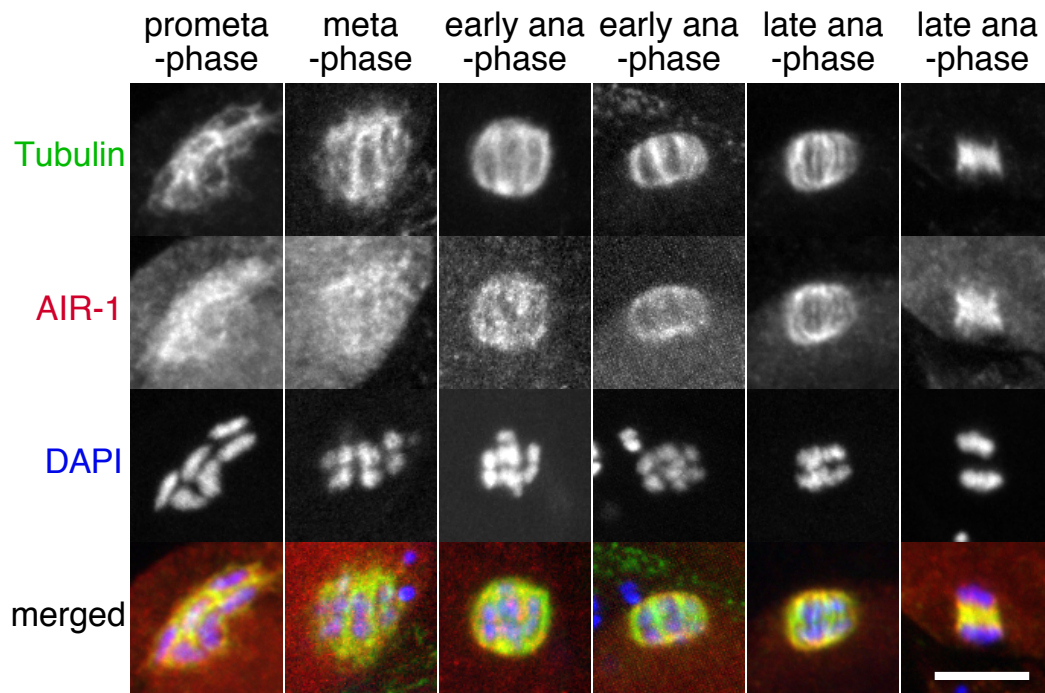
B. Live images of 1-cell mitotic stage embryos expressing GFP:: $\beta$ -tubulin (green), mCherry::Histone (red), and mCherry::AIR-1<sup>K73RT201A</sup> (red). (a) Control, and (b) *air-1n(RNAi);air-1r(RNAi)* (*air-1n*: RNAi for the endogenous *air-1* gene; *air-1r*: RNAi for the RNAi-resistant *air-1* transgene). In (b), the mCherry::AIR-1<sup>K73RT201A</sup> signal is undetectable, and a bipolar spindle is not formed. To visualize weak fluorescence of mCherry::AIR-1<sup>K73RT201A</sup>, contrasts for the red channel is enhanced. Bar=10 $\mu$ m.



**Figure S2. Evaluation of expression level and functionality of the transgene-derived AIR-1 in female meiotic spindle**

A. Western blotting showing the expression of endogenous and transgene-derived AIR-1. (Left) a strain without an AIR-1 transgene, (center) a strain expressing AIR-1<sup>K73RT201A</sup> along with the endogenous AIR-1, and (right) a strain expressing GFP::AIR-1 along with the endogenous AIR-1.

B. RNA-resistant GFP::AIR-1 rescues the phenotype of the endogenous AIR-1 depletion. *in utero* live images of female meiotic spindles in an endogenous-AIR-1-depleted (*air-1n(RNAi)*) embryo expressing mCherry:: $\beta$ -tubulin (green) and RNA-resistant GFP::AIR-1 (red). (a) metaphase, and (b) telophase. The morphology of the female meiotic spindle was normal. Bar=5 $\mu$ m.



**Figure S3. Localization of AIR-1 during meiosis I**

Female meiotic spindles at meiosis I stained with an anti-AIR-1 antibody (red) and an anti-tubulin antibody (green) along with DAPI (blue). Bar=5 $\mu$ m.