Supplemental Information for:

The endo-siRNA Pathway is Required for Robust Patterning During Early Development of *Drosophila*

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Supplemental Figures:

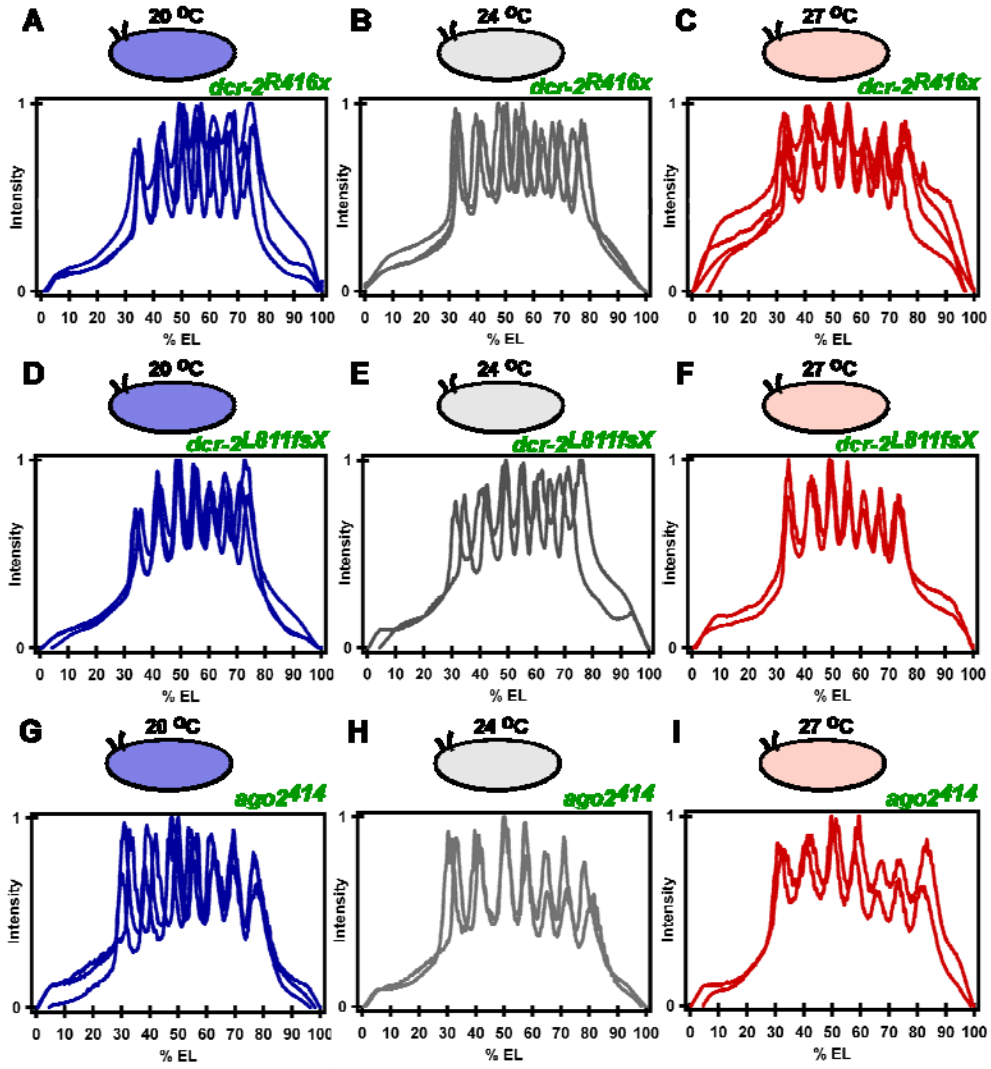

Figure S1. Expression pattern of Even-skipped (Eve) in *dcr-2*^{R416x}, *dcr-2*^{L811fsX}, and *ago2*^{414} mutant embryos developed at a uniform temperature. (A-C) Eve expression is normal in *dcr-2*^{R416x} embryos allowed to develop at a uniform temperature of 20 °C (A), 24 °C (B), or 27 °C (C). (D-F) Eve expression is normal in *dcr-2*^{L811fsX} embryos allowed to develop at a uniform temperature of 20 °C (D), 24 °C (E), or 27 °C (F). (G-I) Eve expression is normal in *ago2*^{414} embryos allowed to develop at a uniform temperature of 20 °C (G), 24 °C (H), or 27 °C (I).

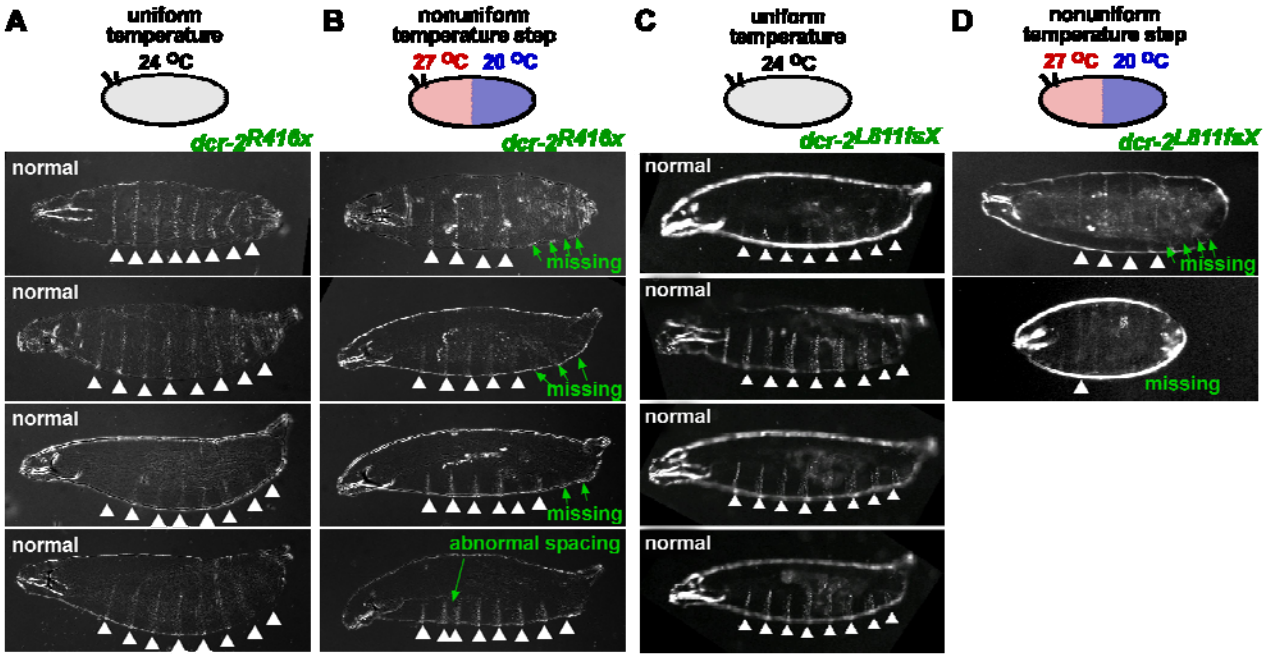

Figure S2. Cuticle preparations of *dcr-2*^{R416x} and *dcr-2*^{L811fsX} larvae developed at uniform temperature (24 °C) or from embryos developed in a temperature step for the first 200 minutes of development and then allowed to reach larval stage at uniform temperature (24 °C). (A,C) All five *dcr-2*^{R416x} and all five *dcr-2*^{L811fsX} larvae that developed at 24 °C appear normal. (B,D) All five *dcr-2*^{R416x} larvae and all three *dcr-2*^{L811fsX} larvae from embryos that were exposed to the temperature step appear abnormal. (B) In *dcr-2*^{R416x} mutants, four of five larvae had between four to six denticle belts. One larva had eight denticle belts, but with abnormal spacing between belts two and three. (D) In *dcr-2*^{L811fsX} mutants, two of three larvae hatched but had only four denticle belts. One larva failed to hatch and had only one clear belt. Total larvae numbers include larvae shown in the main text.

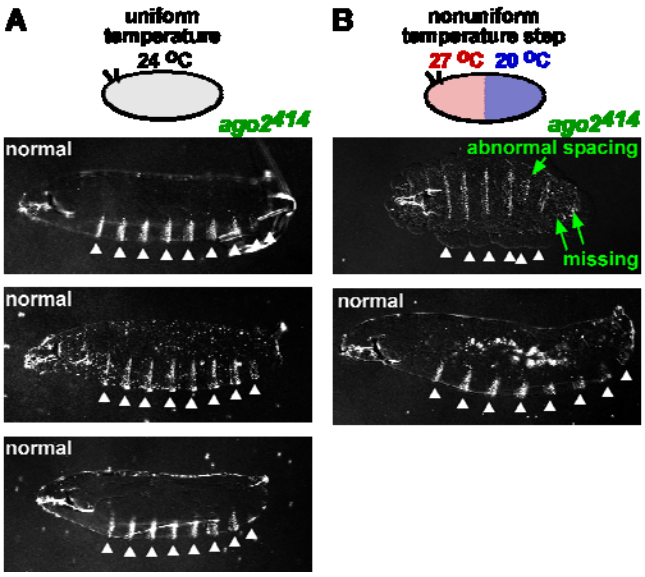

Figure S3. Cuticle preparations of *ago2*^{414} larvae developed at uniform temperature (24 °C) or from embryos developed in a temperature step for the first 200 minutes of development and then allowed to reach larval stage at uniform temperature (24 °C). (A) All four *ago2*^{414} larvae developed at 24 °C appear normal. (B) Two out of three *ago2*^{414} larvae from embryos that were exposed to the temperature step appear abnormal, having four or six denticle belts. One out of three larvae had all eight denticle belts. Total larvae numbers include larvae shown in the main text.

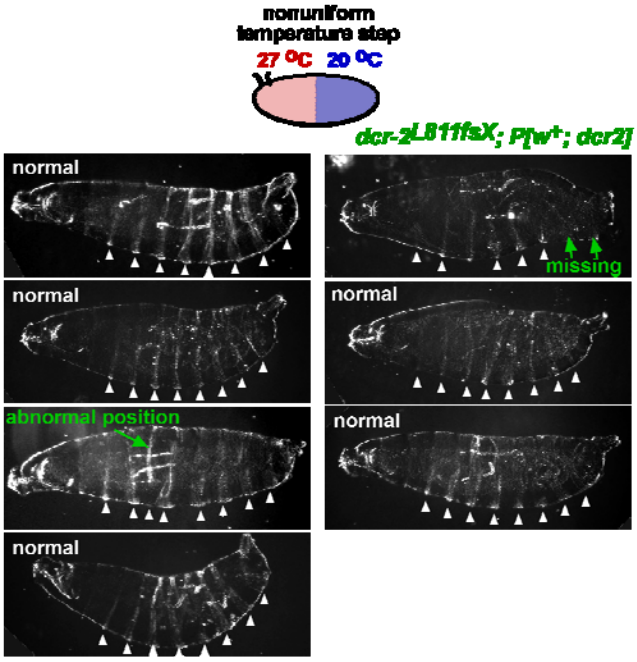

Figure S4. Cuticles of *dcr-2*^{L811fsX}*; P[w*$^{+}$*; dcr-2]* embryos that were developed in a temperature step for 200 minutes and then allowed to grow to larval stage at uniform 24 ^{o}C. Six out of eight larvae developed normally. One larva was missing two denticle belts, and the other larvae had abnormal position of one denticle belt. Total larvae numbers include larvae shown in the main text.

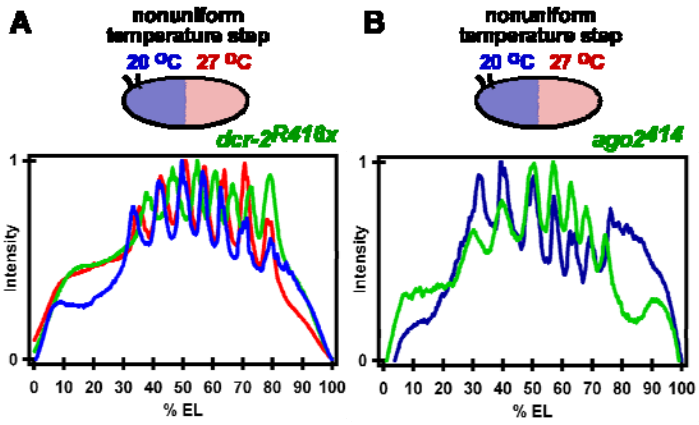

Figure S5. Expression pattern of Even-skipped (Eve) in *dcr-2*^{R416x} and *ago2*^{414} mutant embryos developed in a temperature step with anterior at 20 ^{o}C and posterior at 27 ^{o}C. (A) All three *dcr-2*^{R416x} embryos had the correct number but of Eve stripes, but one *dcr-2*^{R416x} embryo had slightly abnormal position of Eve stripes. (B) Both *ago2*^{414} embryos had the correct number of Eve stripes. These results suggest polarity to the robustness.

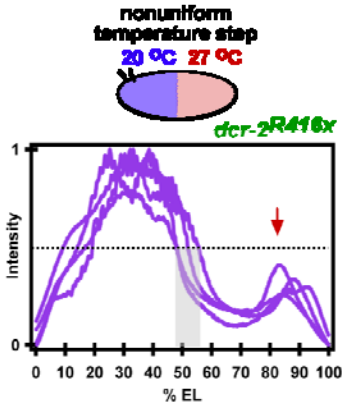

Figure S6. A normal expression pattern of Hunchback (Hb) is observed in *dcr-2*^{R416x} mutant embryos developed in a temperature step with anterior at 20 ^{o}C and posterior at 27 ^{o}C.

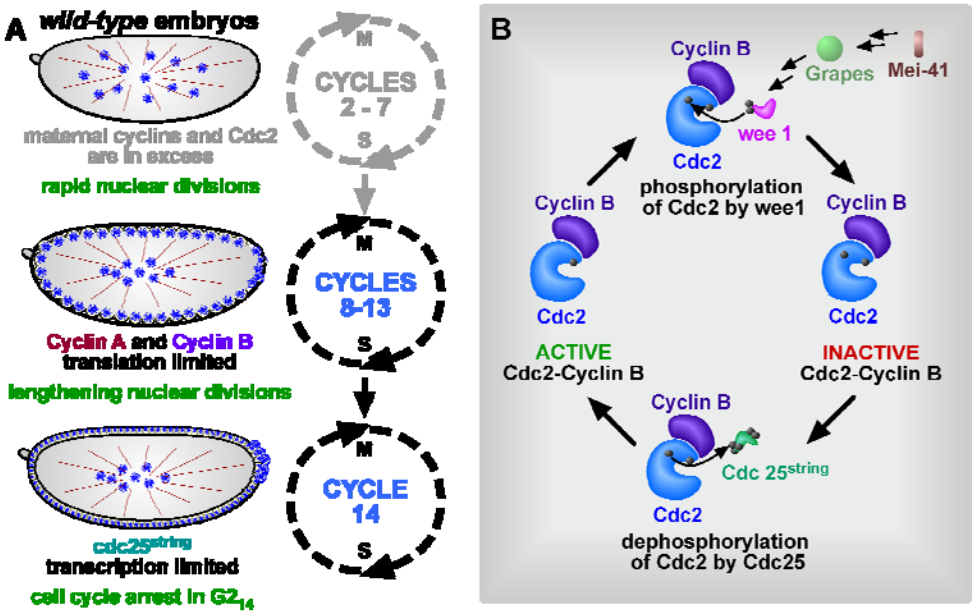

Figure S7. Regulation of cycles 2-14 in the *Drosophila* embryo. (A) During cycles 2-7, all maternal cyclins and Cdc2 are in excess. The nuclear divisions proceed rapidly, and are not limited by the concentration of cyclins or Cdc2. During nuclear division cycles 8-13, Cyclin A and Cyclin B are degraded, presumably by an increase in a nuclear factor. The translation of additional Cyclin A and Cyclin B protein becomes rate limiting, and nuclear division cycles lengthen progressively from cycles 8-13. During nuclear division cycle 14, maternal *cdc25*^{string} is degraded, and transcription of zygotic *cdc25*^{string} becomes rate limiting, causing a cell cycle arrest in $G2_{14}$. (B) Molecular components that drive mitosis in the cell cycle.