

**Figure 7: F-actin is largely normal in *exc-6* and *fhod-1* mutants. (A) Widths of F-actin bundles.** The width of basal F-actin bundles in spermathecae do not differ statistically between wild type, *fhod-1(tm2363)*, *exc-6(gk386)* and *fhod-1(tm2363);exc-6(gk386)*. Data shown are the average of two independent experiments (n = 120-125 actin filament bundles from 12-14 spermathecae per strain). **(B-D) Spermathecae of AJM-1::GFP expressing animals were stained with fluorescent phalloidin to mark F-actin.** Maximum intensity projections (MIP) and XZ slices (XZ) reconstructed from confocal Z-stacks are shown. Magnified views of boxed sections of MIPs (B'-D') and XZ slices (B''-D'') are also shown. Examples include junctions (*arrowheads*) with no visible associated F-actin (B-B''), junctions with associated F-actin (C-C''), and junctions with adjacent thickened F-actin bundles at the basal surface (D-D''). Scale bars, 10  $\mu$ m. **(E) Frequency of visible junction-associated F-actin.** Worms with the single or double formin mutations have a slightly higher proportion of junctions with F-actin than wild type. **(F) Frequency of thickened F-actin bundles adjacent to junctions.** Worms with the *exc-6(gk386)* mutation have a higher proportion of junctions with thickened adjacent F-actin bundles than wild type animals, while *fhod-1(tm2363)* mutants have lower, and *fhod-1(tm2363);exc-6(gk386)* double mutants have nearly wild type proportions. (E, F) are each the combined data of four independent experiments. Bars indicate mean values, and n indicates the number of spermathecae analyzed, with 2 to 13 visible junctions scored per spermatheca.

### Supplementary Figure Legends

**Figure S1: RNAi-mediated knockdowns of *fhod-1* and *exc-6* in wild type animals recapitulate brood size deficiencies of *fhod-1(tm2363)* and *exc-6(gk386)* mutants.** RNAi-sensitive worms were subjected to RNAi against *fhod-1* or *exc-6* or both. **(A) Total brood sizes.** Counting eggs over the lifetime of hermaphrodite worms show that *fhod-1(RNAi)* and *exc-6(RNAi)* additively contribute to a reduced brood size. Note, the untreated RNAi sensitive worms used here produce smaller broods than wild type (Fig.1A). **(B) Timing of egg laying.** Counting eggs laid per animal over the indicated time intervals demonstrates that smaller broods of RNAi-treated animals are not due to early cessation of egg laying, but result from a slower rate of egg production. \*\*\* indicates  $p < 0.001$ . n.s. indicates not statistically significant. For all, values are expressed as means, and error bars indicate one standard deviation. Results shown are representative for three independent experiments.

**Figure S2: Ovulation was visualized in live *fhod-1(tm2363);exc-6(gk385)* worms using DIC microscopy.** In all images, (\*) indicates the ovulating oocyte or fertilized embryo, and (SP) indicates the spermatheca. Stages expected for a normal ovulation are: *Pre-ovulation* when the proximal oocyte has an intact nucleus, *Nuclear Envelope Breakdown* (NEBD) and coincident rounding of the proximal oocyte, *In Spermatheca* after ovulation, when the oocyte has entered the spermatheca, and *In Uterus* when the fertilized embryo (\*) has exited the spermatheca to the uterus. In these two examples of *fhod-1(tm2363);exc-6(gk386)* hermaphrodites, we observe (A) *Failed Entry* of the proximal oocyte into the spermatheca following NEBD, and (B) *Breakage* of the proximal oocyte into two pieces (two asterisks) during entry into the spermatheca. Scale bars, 100  $\mu$ m. Time stamps indicate min:sec.

**Figure S3: Requirements for *fhod-1* in brood size.** The effects on brood size of *fhod-1(tm3138)*, predicted to eliminate part of the FHOD-1 DID, and of *fhod-1(tm2363)*, predicted to eliminate the FH2 domain, were compared, as was the ability of *fhod-1::gfp* to rescue mutant defects. **(A) Total brood sizes.** Counting eggs laid over the lifetime of hermaphrodite worms shows that *fhod-1(3138)* does not result in reduced brood size, and that *fhod-1::gfp* does not rescue brood size defects caused by *fhod-1(tm2363)*. \* indicates  $p < 0.05$ , \*\* indicates  $p < 0.01$ , and \*\*\* indicates  $p < 0.001$ . **(B and C) Timing of egg laying.** Counting eggs laid per animal over the indicated time intervals demonstrates that (B) *fhod-1(tm2363)* and *fhod-1(tm2363);fhod-1::gfp* worms lay eggs at similar rates, as do (C) wild type and *fhod-1(tm3138)* mutant worms. Values are expressed as means, and error bars indicate one standard deviation.

**Figure S4: FHOD-1::GFP is not detected in the somatic gonad.** Distal gonads (A, B), proximal gonads (C, D) and spermathecae (E, F) of wild type worms and worms expressing FHOD-1::GFP were stained with fluorescent phalloidin to mark F-actin, and with DAPI to mark DNA. We did not observe any green fluorescence in FHOD-1::GFP-expressing animals above background. Scale bars, 10  $\mu$ m.