Expanded View Figures

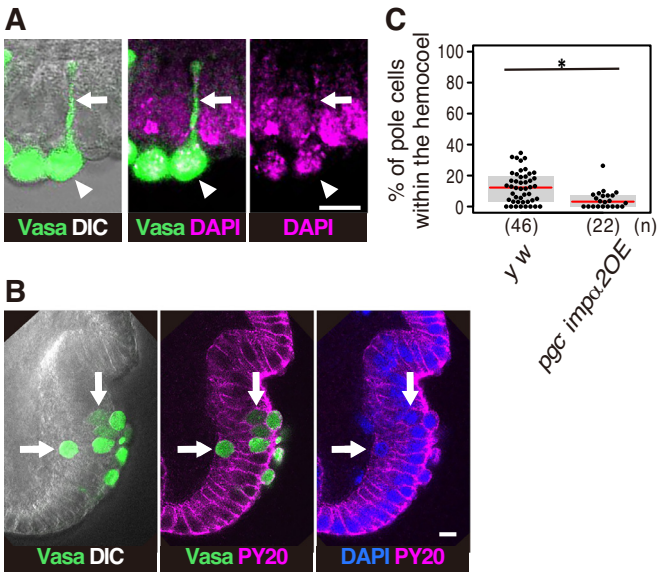

Figure EV1. Phenotypes of *pgc⁻ impα2OE* pole cells.

(**A**) Representative confocal images of pole cell protrusions (arrows) in *pgc⁻ impa2OE* embryos double-stained with anti-Vasa antibody and DAPI at stage 6 ($n = 14$ embryos, biological replicates). A DIC image merged with the Vasa signal is shown on the left. The main body of the pole cell with the nucleus (arrowheads) remained on the outside, but its protrusion penetrated the somatic-cell layer (arrows). The pole cell protrusion (arrows) never reached beyond the somatic-cell layer into the yolk. (**B**) Representative confocal images of posterior-dorsal region of gastrulating *pgc⁻ impa2OE* embryos (stage 6/7) triple-stained with anti-Vasa antibody, PY20 antibody, and DAPI ($n = 22$ embryos, biological replicates). A DIC image merged with Vasa is shown on the left. Arrows indicate pole cells within the epithelium of the midgut primordium. They were intermingled with the epithelial cells in the midgut primordium. In (**A**, **B**) Anterior up, posterior down, ventral left, dorsal right. Scale bars, 10 µm. (**C**) Beeswarm plots showing the percentage of pole cells within the hemocoel in *y w* and *pgc⁻ impa2OE* embryos at stages 7–9. Black dots represent individual embryos (biological replicates). Red lines represent the medians, and the boxes indicate the interquartile range (IQR; 25th–75th percentiles). All data points are shown; therefore, minima and maxima correspond to the lowest and highest dots, and whiskers are not displayed. "*n*" indicates the number of embryos examined per genotype. Statistical significance was assessed using Mann–Whitney *U* test. $^*P < 0.01$ ($P = 0.00098$).