



**Figure 3-figure supplement 2. BiFC interaction in oocytes and testing permeability to  $\text{K}^+$ .** (A) Bimolecular fluorescence complementation (BiFC) analysis of BRUSH and brush fused to either a N-terminal (VN) or C-terminal (VC) fragment of Venus (YFP). Shown are representative oocytes three days after co-injection. The bottom right image of each panel (VN fusion alone) displays the background oocyte fluorescence. (B) Control BiFC experiments utilizing ATKAT1. Images are overlays of brightfield and fluorescent images. (C) Current responses of an oocyte expressing BRUSH-YFP in the presence of 30 mM  $\text{CaCl}_2$ . (D) Currents of a brush-YFP injected oocyte in the presence of 30 mM  $\text{CaCl}_2$  (left) or 60 mM KCl (right). (E) Current-voltage relations of BRUSH-YFP (open triangles,  $n=8$ ), brush-YFP (closed circles,  $n=14$ ), and oocytes injected with water or YFP (closed triangles,  $n=16$ ). (F) Mean relative current-voltage relations ( $n=4$ ) in the presence of 30 mM  $\text{CaCl}_2$  (closed circles) and 60 mM KCl (open circles)  $\pm$  standard deviations. Currents were normalized to the value at -120 mV in the presence of 30 mM  $\text{CaCl}_2$ . Scale bars (A,B) represent 250  $\mu\text{m}$ .