

Figure S1

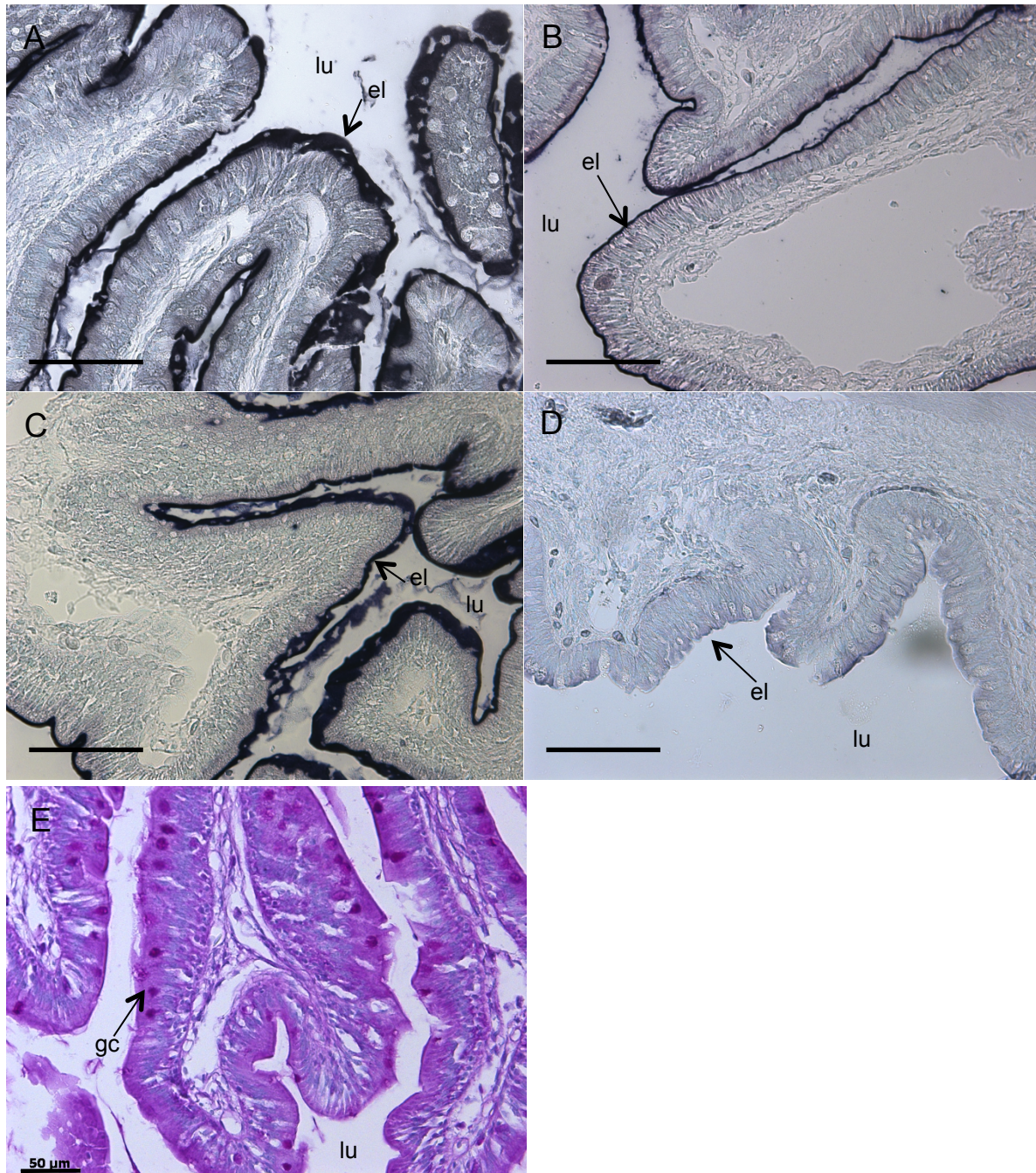


Fig. S1. Histochemical localization of alkaline phosphatase activity and PAS-positive goblet cells in the intestine of fed, fast or refed *X. laevis*. The intestines were collected from frogs that were fed for 22 days (*A and E*), fasted for 22 days (*B*), fasted for 21 days and then refed for 1 day (*C*) and fasted for 5 months (*D*). *A-D*; alkaline phosphatase activity; *E*; PAS-staining. el; epithelial layer; lu, lumen; gc; goblet cells. Bar, 500 μm (*A-D*) and 50 μm (*E*).

Alkaline phosphatase histochemistry was carried out according to the Cox and Singer method (1999). Fixed tissue sections were soaked in 0.2% Tween-20/PBS for 10 min and then rinsed in PBS for 10 min. Sections were incubated with the labeling reaction solution contained 0.17 mg/ml 5-bromo-4-chloro-3-indolyl-phosphate and 0.33 mg/ml nitro blue tetrazolium in reaction buffer (100 mM Tris-HCl, 100 mM NaCl, 5 mM MgCl_2 , pH 9.5) for 30 min in the dark. De-waxed sections were stained with periodic acid-Schiff-hematoxylin (PAS-H) to localize the goblet cells (McManus, 1948).

Cox WG, Singer VL. A High-resolution, fluorescence-based method for localization of endogenous alkaline phosphatase activity. *J Histochem Cytochem.* 1999;47(11):1443–55.

McManus JF. Histological and histochemical uses of periodic acid. *Stain Technol.* 1948;23(3):99-108.