



Supplemental Figure 2: Effect of protein concentration on retinoschisin binding efficiency by **(A)** Hek293 cells or **(B)** Y-79 cells, prepared and treated as described for the ^3H -labeled ouabain binding assay. **(A)** Hek293 transfected with expression constructs for ATP1A3 and ATP1B2 were subjected to 0, 0.5, 1, 1.5, or 2, µg/mL recombinant retinoschisin (RS1) and **(B)** Y-79 cells were subjected to 0, 75, 150, 225, or 300 ng/mL recombinant retinoschisin (RS1). After intensive washing, retinoschisin binding was analyzed by Western blot analyses with antibodies against retinoschisin. ACTB staining served as loading control. Densitometric quantification of retinoschisin binding was performed on immunoblots from four **(A)** or six **(B)** individual experiments. Signals were normalized against ACTB and calibrated against signals for 2 µg/mL **(A)**, or 300 ng/mL **(B)** recombinant retinoschisin. The saturation of retinoschisin binding signals was observed at 1 µg/mL for Hek293 cells and at 75 ng/mL for Y-79 cells.