



SUPPLEMENTAL FIGURE 2. **Standardizing the amounts of injected Keap1 mRNA.** *A*, Experimental scheme. Nrf2 and/or FLAG (*FL*)-tagged Keap1 mRNA were co-injected into embryos at the one-cell stage. After 6 hours, proteins extracted from whole embryos were analyzed by SDS-PAGE. *B*, Immunoblot analysis. Proteins extracted after 6 hours from whole embryos injected with 75 pg of FL-Keap1a or 200 pg of FL-Keap1b were analyzed by SDS-PAGE, followed by immunoblotting with anti-FLAG antibody. *In vitro* synthesized proteins were also analyzed for comparison (*IVT*). The positions of protein mass markers are shown. The *arrowhead* indicates FLAG-tagged Keap1 proteins. *C*, Nrf2-repressing activities of FL-Keap1a and FL-Keap1b. Seventy-five pg of FL-Keap1a or 200 pg of FL-Keap1b mRNAs were co-injected with 40 pg of Nrf2 mRNA into embryos at the one-cell stage. After 8 hours, *gstp1* expression in embryos was analyzed by RT-PCR. *ef1 α* expression was used to standardize the amount of cDNA.