



Supplementary Figure. $H_2^{18}O$ labeling optimization trial. A cohort of laboratory embryos (from adult 5D zebrafish not subjected to dietary manipulation) was incubated with increasing concentrations of $H_2^{18}O$ (0% to 50% v/v in EM, n=2 samples of 15 embryos each, except for 50%, n=1) for 24 h from 48 to 72 hpf to determine the % $H_2^{18}O$ that yielded the greatest label incorporation. Lipid species were identified and quantified as described for lipidomics analyses (See *Methods*) **A.** Incorporation of the label for representative phosphatidylcholine (PC) species are shown; incorporation was measured as the differences in the ratios of $[M+2+H]^+/[M+H]^+$ peak intensities in the $H_2^{18}O$ incubated minus the ratios of $[M+2+H]^+/[M+H]^+$ peak intensities in the H_2O incubated (PeakView software, SCIEX). Incorporation was found to be highest in the 40% v/v $H_2^{18}O$ incubation. **B.** Shown are four representative PC species fitted with non-linear regression curves showing a plateau in label incorporation at 40% v/v $H_2^{18}O$. **C-D.** Incorporation of the $H_2^{18}O$ label in PC with saturated (**C**) and highly unsaturated (**D**) acyl chains; indicating optimal labeling at 40% v/v $H_2^{18}O$ for both.