



**SUPPLEMENTARY FIG. S5.** Analysis of neutralizing antibody (nAb) against the rAAV2/5 capsid after readministration in adult life. An rAAV transduction inhibition assay was performed to analyze nAb against the rAAV2/5 capsid in serum. Samples were analyzed 1 month post-readministration after initial fetal and neonatal rAAV2/5 delivery ( $1.5 \times 10^{10}$  GC of rAAV2/5-fLUC per animal). In addition, adult control mice that received a single vector dose were analyzed in parallel. Serial dilutions of mouse sera were incubated with rAAV2/5-fLUC and tested for inhibition of transduction in HEK293T cells. Transduction was quantified by measuring relative light units per total amount of protein and is expressed as mean percent transduction of no-serum control  $\pm$  SEM. *Abbreviations:* Fetal-R or Neonatal-R, initial fetal or neonatal vector delivery followed by readministration at 3 months and serum collection at 4 months; Adult ctrl, adult control mice that received a single vector dose, serum collection 1 month postinjection.