

Hypothermic Protection in Neocortex is Topographic and Laminar, Seizure Unmitigating, and Partially Rescues Neurons Depleted of RNA Splicing Protein RBFOX3 in Neonatal Hypoxic-Ischemic Piglets

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Supplementary Materials:

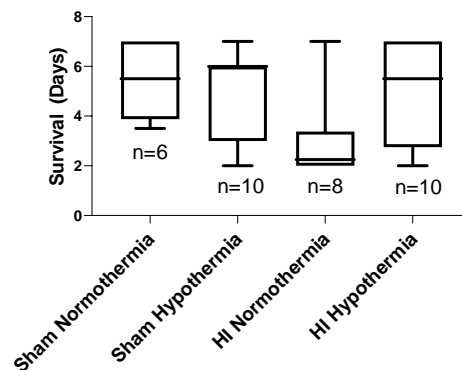
Animal attrition

Forty piglets were anesthetized for intubation and catheter placement. Among piglets randomized to HI and normothermia, one animal died of refractory hypotension during anesthesia and a second piglet could not be extubated. One HI hypothermia piglet had a pulmonary hemorrhage, and another had a cardiac arrest, excluding them from the study. One sham hypothermia piglet could not be intubated and a second did not sustain adequate ventilation upon extubation and thus did not complete the protocol. Three HI-hypothermia, 1 HI-normothermia, and 1 sham-hypothermia piglet received dopamine during anesthesia. Hypoxia-ischemia, HI. Whiskers show the 5-95th percentiles.

Anesthesia toxicity

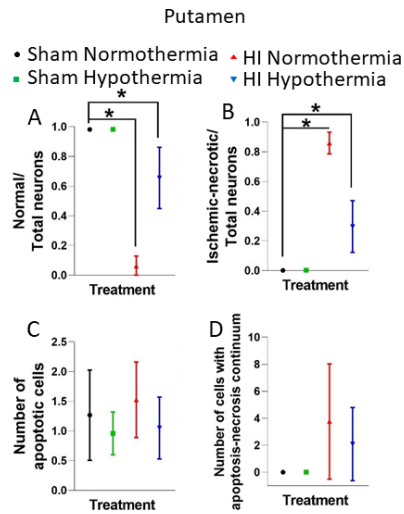
To examine the potential effects of the anesthetic regimen with and without hypothermia, we compared the ratio of normal-to-total neurons among naïve unanesthetized (n=6), sham normothermic (n=6), and sham hypothermic (n=10) piglets. The ratio of normal neurons did not differ between anterior and mid-parietal somatosensory and motor cortices among the unanesthetized and sham procedure piglets ($p>0.05$ for all comparisons).

Supplementary Figure 1



Supplementary Figure 1. The duration of survival did not differ among groups ($p=0.078$).

Supplementary Figure 2. HT protects the putamen from HI injury



Supplementary Figure 2. HT reduced the burden of HI neuronal injury in putamen after adjusting for survival duration. **A.** Loss of normal neurons occurred in HI-HT piglets ($p=0.008$) and HI-NT ($p<0.001$) compared to sham-NT piglets. **B.** More ischemic necrosis occurred in HI-HT ($p=0.014$) and HI-NT ($p<0.001$) piglets compared sham-NT piglets. **C, D.** The number of cells with apoptosis or the apoptosis-necrosis continuum did not differ among groups. Data are shown with means and 95% CIs. * $p<0.05$. Hypoxia-ischemia, HI.

Full Length NeuN western blot

