Isoflurane Exposure in Neonatal C57BL/6 Mice Creates a Concentration Dependent Deficit in Context Pre-exposure Facilitation Effect as Juveniles but Not Adults

Christopher Ward, M.D., Timothy DeYoung, B.S., Gordon Barr, Ph.D., Francis McGowan, M.D., Maryellen Eckenhoff, Ph.D., Roderic Eckenhoff, M.D., Ph.D.

Department of Anesthesiology & Critical Care Medicine, Children’s Hospital of Philadelphia, Philadelphia PA

Background

- Anesthesia is used to facilitate surgical and radiological procedures in millions of children every year, but has repeatedly been shown to cause extensive apoptotic cell death in developing and more recently mature animals.
- Despite overwhelming histological evidence that exposure to anesthesia can cause neuronal cell death, the clinical consequences of this phenomenon remain unknown.
- Previous studies have shown anesthetic exposure to postnatal day (PND) 7 mice caused extensive apoptotic damage and transient deficits in Context Pre-exposure Facilitation Effect (CPFE) as juvenile mice.
- Previous work has also shown a stepwise worsening apoptotic effect from increasing concentrations of isoflurane,[1] but no study has correlated the amount of apoptosis with a concurrent stepwise worsening in a learning/behavioral effect.

Methods

- PND 7 and PND 21 mice (n=74) were either fasted in 30% Oxygen or exposed to isoflurane anesthesia (1% or 1.5%) in 30% oxygen for 6 hours.
- At either PND 28 or PND 49 the animals were subjected to a 3 day CPFE paradigm.
- Day 1 the animal was placed in the context environment for 5 minutes.
- Day 2 the animal was placed in the context, given a shock at 0.5mA for 2 seconds and removed from the context 30 secs later.
- Day 3 the animal was returned to the context and freezing behavior was recorded for 5 minutes.

Results

Control

- PND 7 mouse brain slices demonstrating increasing neocortex staining for Caspase-3 following exposure to 6 hours of control, 1% or 1.5% Isoflurane

Anesthesia Exposure

30% Oxygen

30% Oxygen and 1% Iso

30% Oxygen and 1.5% Iso

- Using t-test analyses, statistical difference for mean freezing score, and total time freezing, but not latency to start of the first freezing episode was observed between the PND 7 exposed animals done at PND 28. No statistical difference was observed at any other exposure age or age of testing.

Discussion

- Retrospective human studies looking at the possible consequences of anesthetic exposure and long term learning and behavior outcomes in children have given inconclusive results and are unable to separate surgical and anesthesia related outcomes.
- These results suggest a transient deficit that did not persist into adulthood in which the magnitude of the deficit correlates with the amount of anesthesia the animal was exposed. This mirrors clinical retrospective studies that demonstrated links with multiple general anesthetic exposures or total duration of the exposure to neurocognitive deficits later in life.
- Given the rodents’ ability to recover later in life, the clinical consequences of this neuronal destruction may be transient in nature and the human brain potentially can overcome anesthetic insult incurred during any window of vulnerability.
- Translating the combined results of different rodent testing paradigms and CPFE in particular, any clinical deficit or behavior change that may be caused in our pediatric population may likely be small and transient and need a very specific and sensitive testing battery to detect.
- The results of this study also suggest that the total amount of anesthesia and time under anesthesia should be limited to the best of our clinical abilities to keep our patients safe while under our care and beyond.

References