Surgical Stress-Induced Glutamate Dysregulation in Prefrontal Cortex of Neonatal Piglets

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ABSTRACT

Apoptotic neurodegeneration after anesthesia is a well-established phenomenon in animal models. However, the effects of surgical stress on the developing brain remain poorly understood. Glutamate activity in the brain is known to be dysregulated in the setting of major physiologic stress, both physical and psychological. Further, glutamate dysregulation has been shown to activate signaling pathways that lead to apoptosis. Anesthesia-induced imbalances in GABA and glutamate activity combined with glutamate dysregulation caused by major surgical stress may lead to significant dysfunction in the developing brain. We hypothesized that major surgical stress would lead to glutamate dysregulation in the developing brain of neonatal piglets.

Methods:

- 6 piglets: sevoflurane alone (n=3) or sevoflurane + femoral osteotomy (n=3).
- Animals were anesthetized and prepared as previously described by our laboratory
- Enzyme-based microelectrode arrays (MEAs) were used to continuously measure glutamate in the prefrontal cortex of piglets
- Control animals received sevoflurane at 1 MAC for 3h.
- Animals in the surgery group underwent open femoral osteotomy, after which glutamate was recorded for 3h.
- Animals were sacrificed and brain tissue was assessed for proper electrode placement.

Results:

- Sevoflurane-only animals exhibited glutamate concentrations consistent with prior results in animal models
- Piglets that underwent femoral osteotomy showed a consistently higher number of glutamate peaks when compared to anesthesia only animals, but the amplitude of glutamate peaks was more variable
- Glutamate clearance (t80) was highly variable
- When comparing the acute change (15 minutes post-surgery), glutamate concentrations in the sevoflurane only group were significantly lower than those in the osteotomy group at 5 minutes (8.33 ± 2.31 μM vs. 30.0 ± 16.3 μM, P<0.0001), 10 minutes (8.05 ± 2.38 μM vs. 27.2 ± 14.0 μM, P<0.0001), and 15 minutes (7.73 ± 2.59 μM vs. 26.1 ± 14.0 μM, P<0.0001).

Conclusion: Our data support the hypothesis that an acute change in glutamate concentration and activity occurs immediately after major surgical stress when compared to animals that did not receive surgical stress.

RESULTS

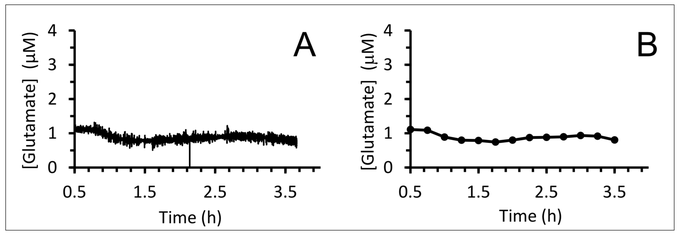


Figure 1. Glutamate concentration during a 3h sevoflurane exposure. Raw data shown (Panel A) and smoothed data averaged in 15 minute bins (Panel B). Data shown for 1 animal.

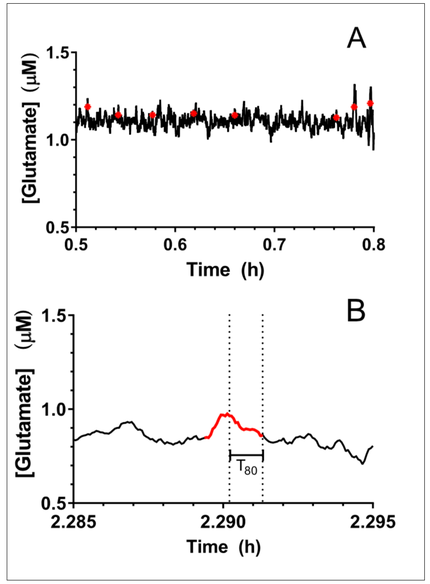


Figure 2. Red dots represent glutamate peaks where the S:N ratio ≥ 3 (panel A). Peak decay (t80), representing time taken to restore glutamate concentration to baseline, was highly variable. For a 3h sevoflurane exposure, t80 was 4.68 ± 0.82 s (averaged over 3h).

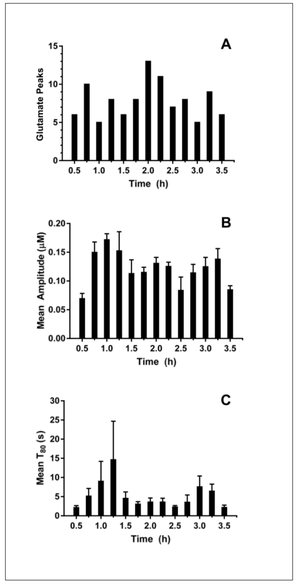


Figure 3. Glutamate peaks (Panel A), mean peak amplitude (Panel B) , and mean t80 in 30 minute bins over the course of a 3h exposure to sevoflurane.

RESULTS

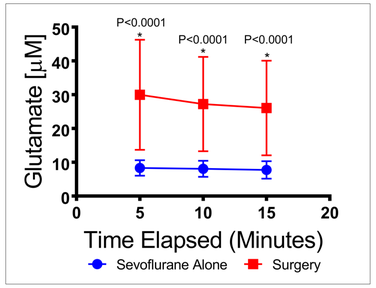


Figure 4. Glutamate concentration is increased at 5, 10, and 15 min post-osteotomy when compared to animals that received anesthesia alone. Data are displayed as mean ± SD.

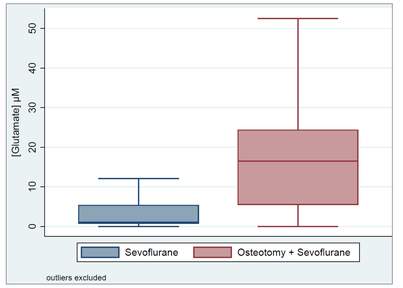


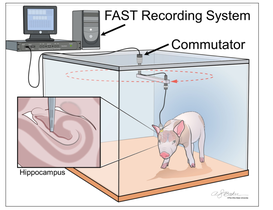
Figure 5. Average of all glutamate measurements over a 3h period during sevoflurane alone vs. sevo + femoral osteotomy.

CONCLUSIONS

- Data support our hypothesis that extracellular glutamate is increased in the setting of major surgical stress when compared to sevoflurane alone
- The significance of this finding remains unclear
- Future experiments will focus on the mechanism(s) of glutamate dysregulation and the downstream, potentially toxic effects (apoptosis)
- Our study is limited by its acute nature and short time course
- Surgical stress may have a long-lasting effect on glutamate dysregulation, leading to excitotoxicity and potentially neurocognitive defects

FUTURE DIRECTIONS

- Determination of the long-term effect of surgical stress on glutamate dynamics and the resultant behavioral changes is crucial
- Measurement of glutamate concentration in awake, freely moving animals is planned



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