The Intra-amniotic Administration- an Emerging Method to Investigate Necrotizing Enterocolitis, In Vivo (Gallus gallus)

Nikolai Kolba, Cydney Jackson, and Elad Tako

Cornell University

Objectives: Demonstrate necrotizing enterocolitis (NEC) in a novel in vivo (*Gallus gallus*) model through intra-amniotic administration of dextran sodium sulfate (DSS, demonstrated agent to induce NEC), to induce NEC pathology, pathophysiology, and negative bacterial-host interactions within a premature gastrointestinal system.

Methods: Gallus gallus embryos were injected intra-amniotically with 1 mL DSS in DI H_2O. Four treatment groups (0.1%, 0.25%, 0.5%, and 0.75% DSS) and two controls (H_2O/non-injected controls) were administered. Upon hatch, blood, cecum, small intestine, and liver were collected to assess hemoglobin and intestinal permeability, intestinal microbiota alterations, intestinal morphometric assessment, and mRNA gene expression of relevant key nutrient transporters/enzymes inflammatory proteins, respectively.

Results: Results indicated that intestinal permeability was significantly increased post DSS exposure, and negative intestinal morphological changes were found. In the 0.50% and 0.75% DSS groups, villus surface area and goblet cell diameter have significantly decreased ($p < 0.05$). Furthermore, there was a significant ($p < 0.05$) increase in pathogenic bacterial abundance (*E. coli* and *Klebsiella*) in the 0.75% DSS group compared to the controls, demonstrating cecal microbiota dysbiosis. Taken together, these results demonstrate striking similarities between current observations in the *Gallus gallus* model compared with NEC patients.

Conclusions: This study is the first to demonstrate NEC symptoms through intra-amniotic administration of DSS in vivo (*Gallus gallus)*, whereas previous studies have utilized rodent and pig models. The results of this study are promising evidence to investigate increased concentrations of DSS further to cause more severe NEC symptoms and identify potential novel biomarkers of less severe NEC cases. The development of such a model also allows the assessment of potential dietary bioactives that may ameliorate the effects of NEC.

Funding Sources: No external funding was utilized for the study.