PERINATAL ACETAMINOPHEN TOXICITY IS MEDIATED BY CYP2E1 IN A TIME AND CELL TYPE SPECIFIC MANNER

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Purpose of the Study

Acetaminophen (APAP) exposures occur in 50-60% of pregnancies in the US[1] and is concerningly associated with childhood respiratory morbidity[2–23] . The mechanism behind this remains unknown. Toxicity occurs from cell-specific expression of *Cyp2e1,* the enzyme responsible for metabolizing APAP into the mitochondrial toxin N-acetyl-para-benzo-quinone (NAPQI). In the developing murine lung, prenatal pulmonary *Cyp2e1* expression peaks during the saccular stage (E17.5-P4) and is limited to the myofibroblast. We hypothesize this peak in *Cyp2e1* expression predicts susceptibility to APAP-induced lung injury during this critical developmental period.

Methods Used

To confirm dynamic *Cyp2e1* expression in the developing murine lung, RNA was isolated from lungs of wild-type mice from E12-P7 and assessed for *Cyp2e1* expression. CYP2E1 protein was characterized by western blot. To determine a cell-specific pattern, *Cyp2e1* expression in Pdgfrα-GFP labeled pulmonary myofibroblasts was compared to *Cyp2e1* expression in all other lung cell types.
C57BL/6 murine dams (n= 8-16 per condition) were exposed to APAP (250mg/kg IP; 6h) on embryonic day 17 (E17) or 18 (E18). We interrogated a pulmonary transcriptional response in inflammatory (*Il6* and *mmp9*), oxidative stress (*Gclc*, *Hmox1*, *Nqo1*) and apoptotic related factors (*Trp53*, *Puma*, *Noxa*) by qPCR.

Summary of Results

In the time interval we analyzed, *Cyp2e1* expression was low in the developing mouse lung until E18 when it abruptly peaks. CYP2E1 protein was detected by western blot at both e17 and e18. *Cyp2e1* expression at E18 was enriched in Pdgfraα–GFP positive lung myofibroblast cells. Following APAP treatment of pregnant dams at E17 and E18, *Cyp2e1* expression is increased ($p<0.05$). In E18 APAP exposed embryos we observed increased expression of oxidative stress genes: *Gclc*, *Hmox1* and *Nqo1* and induction in p53 mediated apoptotic genes: *Puma* and *Noxa,* and in the inflammatory response gene *Mmp9* ($p<0.05$).

Conclusions

Using a murine model we demonstrated that pulmonary *Cyp2e1* expression is timing and cell type-specific, peaking at E18 and limited to the mesenchymal myofibroblast. We also found that a non-lethal dose of APAP resulted in upregulation of expression of genes associated with antioxidant response elements, apoptosis, and inflammation. Continued work is needed to determine whether perinatal APAP exposure has detrimental effects on the developing lung, its function, and the role of pulmonary *Cyp2e*1 in this mechanism of lung injury.