0.72 days; p = 0.02). Subset analyses within the Caucasian population demonstrated continued value for ADI predicting longer LOS (mean LOS 1.1 vs 0.77; p = 0.02) and ED utilization (6% vs 2.4%; p = 0.04).

**CONCLUSION:** Community-level deprivation indices, which transcend race, correlate with post-appendectomy healthcare utilization and provide a novel tool for future quality improvement initiatives to improve care for children living in disadvantaged communities.

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**Prenatal Administration of Heparin Binding Epidermal-Like Growth Factor as a Preventative Strategy for Necrotizing Enterocolitis**

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**INTRODUCTION:** Necrotizing enterocolitis (NEC) is the most common gastrointestinal emergency in neonates, particularly in premature infants. Prenatally administered heparin binding epidermal-like growth factor (HB-EGF) preserves intestinal mucosa, decreasing the incidence and severity of NEC. The objective was to show maternal intraperitoneal (IP) HB-EGF injection is delivered to intestinal tract of rat pups, protecting against NEC by preserving gut barrier function, by measurement of gut permeability and amniotic fluid concentrations.

**METHODS:** Pregnant rats received HB-EGF (800 μg/kg) by IP injection 2 hours before cesarean section at 21 days’ gestation. Three IP injection and 2 control animals were exposed to the NEC protocol, for a total of 5 pregnant female rats with 65 pups. Amniotic fluid collected at cesarean section was assayed with an HB-EGF-specific ELISA (ThermoFisher Scientific) for concentration quantification using linear regression analysis. Gut barrier function was investigated with fluorescein isothiocyanate-labeled dextran, given to pups orogastrically 4 hours before sacrifice. Serum levels measured at sacrifice were compared with time-matched NEC control animals 0, 24, and 48 hours after delivery (p < 0.05).

**RESULTS:** The amniotic fluid concentration in animals receiving HB-EGF had levels 3-fold higher compared with control animals 136.5 vs 52.6. Prenatal HB-EGF treatment decreased fluorescein isothiocyanate-labeled dextran, given to pups orogastrically 4 hours before sacrifice. Serum levels measured at sacrifice were compared with time-matched NEC control animals 0, 24, and 48 hours after delivery (p < 0.05).

**CONCLUSION:** Prenatal IP injection is an adequate route to deliver HB-EGF, and preserved gut barrier function. Maternal administration of HB-EGF might be effective prophylaxis against NEC by preserving gut barrier function.

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**PROM1⁺ Biliary Progenitor Cells Reside in Peribiliary Glands and Proliferate into Cholangiocytes in Response to Cholestatic Injury in Murine Biliary Atresia Models**

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**INTRODUCTION:** Biliary atresia (BA) is a congenital oblitative extrahepatic bile duct (EHBD) injury. In BA, intrahepatic PROM1⁺ hepatic progenitor cells (HPCs) give rise cholangiocytes comprising ductular reactions. Hypothesis: PROM1⁺ HPCs, present in peribiliary glands (PBGs) outside the EHBD lumen, differentiate into cholangiocytes after injury.

**METHODS:** EHBD immunofluorescence was performed in neonatal wild-type (WT) and Prom1 knockout (KO) BalbC mice with rhesus rotavirus (RRV)-mediated BA and Prom1<sup>cre-<br/>er<sup>2.</sup>Rosa26Gfp<sup>(Prom1-Gfp)</sup></sup> B57BL/6 adult mice, wherein GFP⁺ denotes Prom1-expressing cell lineage, after bile duct ligation (BDL). Epithelial organoids from Prom1<sup>cre-<br/>er<sup>2</sup></sup>,Rosa26<sup>tm1TmG</sup> WT and KO EHBDs were grown in Matrigel for 9 days. Confocal imaging was analyzed using ImageJ and Arivis Vision4D. Unpaired t-tests were performed for statistical analyses (p < 0.05).

**RESULTS:** At baseline, Prom1-Gfp mice expression was localized mostly to PBGs. After BDL, GFP⁺ cell lineage replaced nearly the entire EHBD lumen (Fig.). A similar pattern was observed in RRV-mediated BA. PBGs of RRV-treated KO mice were larger than WT PBGs (1,484.4 ± 1,156 vs 126.1 ± 86.3; p = 0.002). After 9 days in vitro, KO EHBD organoids were larger than WT (0.30 ± 0.87 vs 0.09 ± 0.20; p = 0.02), similar to in vivo findings. All organoids were mG⁺ (green), not mT⁺ (red), suggesting that only HPCs gave rise to viable organoids, even with Prom1 KO.
CONCLUSION: PROM1+ HPCs in PBG’s give rise to EHBD cholangiocytes after BDL and RRV-mediated BA. Larger PBGs in RRV KO mice suggests a cholangiocyte differentiation defect, paralleled in vitro with larger organoids. We conclude that PROM1 HPCs promote EHBD epithelial repair; this process might be disrupted in BA.

Figure. Lineage tracing of Tamoxifen-induced GFP expression in Prom1+ HPCs and their progeny in Prom1-Gfp EHBD after BDL.*-Peribiliary Glands (PBGs).

RAS Inhibition Delays Neuroblastoma Tumor Growth and Genes Upregulated in Tumors with High Ras Expression Can Be Identified Using Gene Set Enrichment Analysis

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INTRODUCTION: Despite low incidence of RAS mutations in neuroblastoma, elevated RAS activity is emerging as critical for tumorigenesis due to mutations in upstream activators of RAS or loss-of-function mutations in negative regulators. We developed RAS monobody, NS1, which prevents RAS activation of downstream effectors by inhibiting H-RAS- and K-RAS-mediated signaling. We hypothesized that NS1 monobody induction in orthotopic neuroblastoma mouse model delays tumor formation and growth and that RAS-associated gene signature in MYCN-nonamplified tumors can be identified by unbiased analysis of RNA-sequencing patient cohort.

METHODS: SK-N-FI neuroblastoma cells were stably infected with lentivirus encoded with doxycycline-regulated NS1 expression (SK-N-FI NS1). RAS activity suppression in SK-N-FI NS1 was determined by Western blot. SK-N-FI NS1 cells were injected into the left adrenal gland of immunocompromised mice to generate orthotopic xenografts. Mice received water ± doxycycline every other day and tumor growth was tracked with ultrasound. We used R2 platform to analyze Neuroblastoma-SEQC-498-seqcnb1-dataset (498 patients) and performed gene set enrichment analysis (GSEA) for MYCN-nonamplified, high HRAS expression tumors.