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Basic Science

Abstract Title
Influence of Amniotic Fluid Composition on Gene Expression of Airway Epithelial Cells

Background
Amniotic fluid (AF) plays a vital role in fetal lung development by providing a distending pressure. We have previously shown that AF is also rich in cytokines and chemokines, which through fetal breathing movements, can enter the lung and interact with the airway epithelial cells. Additionally, the profile of these mediators can be influenced by the maternal environment.

Objective
The objective of my study was to determine whether AF cytokines can influence gene abundance in airway epithelial cells.

Methods
AF samples (n = 32) were collected from term, uncomplicated pregnancies undergoing cesarean delivery. Airway epithelial cells, HBEC3-KT, were cultured under liquid-covered conditions and exposed to different AF samples diluted to 50% for 4 hours. RNA was extracted and gene abundance analyzed using the NanoString nCounter Host Response Panel, targeting 744 innate immune–related genes. Correlation heatmaps were generated to visualize associations between AF cytokines and gene abundance.

Results
Correlation heatmap analysis revealed two clusters of correlated AF cytokines that opposingly correlated with the same set of HBEC3-KT genes. Cytokine cluster 1, which included several interferons, was associated with increased abundance this gene set, whereas cytokine cluster 3, which included several platelet derived growth factors, was associated with decreased abundance of the same gene set. For example, protein kinase R (or E1F2AK2) abundance in HBEC3-KT was positively correlated with AF IFNα2 ($R^2 = 0.36$, $p = 2.94 \times 10^{-4}$) and negatively correlated with AF PDGF.AB/BB ($R^2 = 0.16$, $p = 2.35 \times 10^{-2}$).

Conclusion
AF cytokines and chemokines can influence epithelial cell gene expression, with potential implications for early-life lung disease. The opposing regulation of IFNa2 and PDGF.AB/BB demonstrates how distinct proteins can drive different responses on the same genes. Higher IFNa2 with lower PDGF.AB/BB levels in AF may increase specific gene abundance, indicating AF's role in shaping fetal lung development.

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