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Title: *Development of the preterm infant gut microbiome: Impact of nutrition and feeding type*

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Background: Preterm birth, defined as childbirth occurring at less than 37 completed weeks, is a major determinant of neonatal mortality and morbidity and has long-term adverse consequences for health. The human microbiome is the collective genome of the immense ecosystem of trillions of microbes and is considered to have a coevolved relationship with the host. Little is known about the developmental of the gut microbiome in preterm infants and the impact of neonatal nutrition on its development.

Research Hypothesis: Preterm infants are at risk for marked dysbiosis of the gut microbiome due to multiple reasons, including physiological immaturity, prenatal and postnatal factors which may influences the development of a normal gut flora

Aims: To characterize the development of the microbiome in premature infants.

Methods: Infants born in Rambam healthcare campus in Haifa, Israel, were recruited to the study. During hospital stay, blood and fecal were collected from the premature infants. Fecal samples were collected form the parents. Breast milk samples were collected from the mothers. Infants were followed up for one year. Stool samples were analysed using shotgun metagenomic sequencing. Relative abundances of microbial taxa were calculated at the phylum level, according to the belonging of each bacterial species.

Results: 100 premature infants were recruited to the study between 01/2019 and 02/2021, including 51 boys and 49 girls. 34 Infants were born by vaginal delivery and 66 were born by cesarean section delivery. Mean gestational age was 31.55 ± 2.5 weeks (median 32, IQR 29.4- 34). Mean weight was 1584.9 ± 457.5 grams (median 1610, IQR 1229- 1931) The cohort included 51 infants born in singleton deliveries, 20 pairs of twins and 3 triplets. Analyses of shotgun metagenomic sequencing of the first 107 stool samples, which originates from 21 infants, revealed that microbiome diversity increased with time in all infants. A similar trend was observed in almost all infants, with initial samples containing *Proteobacteria* and *Firmicutes* phylum, while *Bacteroidetes* and *Verrucomicrobia* phylum were detected in samples obtained later in the course of the infant's life.

Discussion: Our findings shed light on the early development of the microbiome in premature infants.

Conclusions: An overall similar development pattern was observed in the majority of premature infants. Data collected in this study will be further analysed to study the complex interactions between nutrition and the microbiome.

Key words (U_p to 5): Premature infants, Preterm infants, Metagenomic sequencing, Microbiome diversity, Microbiome development



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Publications associated with the project (PubMed Format): N/A