**Supplemental Material and Methods**

**Fecal samples and general information collection**

Fecal samples of neonates' first defecation after hospitalization were collected by stool collection kit provided by Genesky Biotechnologies Inc., Shanghai, 201315 (China) and immediately frozen in ice boxes. The samples were transported to the laboratory within 30min and stored at −80°C.

**DNA extraction and high-throughput 16S rDNA gene sequencing**

16S rDNA amplicon sequencing was performed by Genesky Biotechnologies Inc., Shanghai, 201315 (China). Using the QIAamp Fast DNA Stool Mini Kit (QIAGEN ART.NO.56104), extracted total genomic DNA. Detecting the integrity of genomic DNA through agarose gel electrophoresis and the concentration and purity of genomic DNA were detected through the Nanodrop 2000 and Qubit 3.0 Spectrophotometer. Amplifying the V4–V5 hypervariable regions of the 16S rDNA gene with the primers 515F (5′-GTGCCAGCMGCCGCGG-3′) and 907R (5′-CCGTCAATTCMTTTR AGTTT-3′)(1) and then sequenced using Illumina NovaSeq 6000 platform (2). Depositing the sequencing data to NCBI's Sequencing Read Archive with the accession ID PRJNA926124.

**Gut microbial analysis**

The raw read sequences were further filtered to remove adapter sequences, the primers, and low-quality reads by QIIME2 (3) and the cutadapt plugin to improve the accuracy of later analysis. Clustering the filtered sequences into operational taxonomic units (OTUs), which ≥97% similarity, the sequence with the highest abundance was considered representative within each cluster (4). Using the species accumulation curve of the sample analyzed with QIIME2 to assess the rationality of sample content. We evaluated Alpha diversity using abundance indices and diversity indices. Chao 1 and ACE represent abundance, and Shannon and Simpson represent diversity. Venn diagram, visualized with QIIME2 based on OTUs abundance, was used to show the groups' richness and similarity of gut microbiota composition. Principal component analysis (PCA) in Beta diversity (based on Bray–Curtis distance calculated with QIIME2) was used to evaluate community composition and structure of gut microbiota.

Through the linear discriminant analysis (LDA) histogram and the cladogram, Lefse analysis (5) was used to obtain species with significant differences in abundance between the LOS group, the pneumonia group, and the control group. We used the differences in the relative abundance of gut microbiota at phylum and genus levels to evaluate the differences in gut microbiota composition between the LOS, pneumonia, and control groups. We used the ROC curve calculated and displayed with R software (Version 4.2.2) to assess effective biomarkers for LOS. We used Principal component analysis (6) based on the phylum and genus levels of gut microbiota to evaluate the value of their contribution to LOS and pneumonia.

**Statistical analysis**

  SPSS26 and R software (Version 4.2.2) analyzed the experimental data. For measurement data, if they were normal distribution (e.g., maternal age, Gestational age, Birth weight), they were expressed as mean±SD (X±S) and used independent sample t-test for comparison difference between the two groups; Use the median and interquartile range [M (P25, P75)] express non-normal distribution (e.g., Age, WBC, Plt, CRP, PCT, Alpha diversity analysis index, phylum, and genus levels relative abundance). Use the rank sum test to compare the difference between the two groups. Use percentages express Categorical data, and the chi-square test evaluated the comparison differences between the two groups. They considered the value of p<0.05 to be statistically significant.

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