Extraintestinal pathogenic *Escherichia coli* (ExPEC) are responsible for various infections outside the gastrointestinal tract in humans, farm animals, and pets. Many studies show that ExPECs share several virulence factors, despite having been isolated from different hosts and niches. Neonatal meningitis *E. coli* (NMEC), one of the ExPEC sub-pathotypes, has emerged as the major cause of neonatal meningitis among premature neonates with very low birth weight (<1.5 kg). However, there is only limited information on NMEC epidemiology, particularly in Brazil. Our goal is to assemble and characterize a collection of Brazilian NMEC strains. In this work we sequenced the whole genome and characterized 12 NMEC strains obtained from liquor, 11 isolated in hospitals of São Paulo city between 1985 and 1990, and 1 strain isolated in a hospital of Espírito Santo in 2018. The clonality status of the 12 strains was verified by PFGE; 2 strains displayed the same PFGE pattern, and thus 11 strains were sent for sequencing of the whole genomes using Illumina (MicrobesNG company). The genomes were than analysed *in silico* for their phylogenetic status, serotype, multilocus sequence type (MLST) and virulence genes associated with ExPEC pathotype. Of the 12 strains obtained so far, 3 belonged to the O1:K1:H7 serotype, 3 to O6:K1:H31, 2 to O7:K1:H- strains, 1 to O7:K1:H45, 1 to -:H33 strain Chapman ([https://pubmlst.org/escherichia/](https://pubmlst.org/escherichia/)) revealed 3 ST59 strains, 3 ST62, 3 ST127, 1 ST48, 1 ST93, and 1 unclassified strain. We checked the ECOR phylogenetic group *in silico* using the new Clermont method ([http://clermontyping.iame-research.center/](http://clermontyping.iame-research.center/)); while available epidemiological studies on NMEC show that they fall mainly in phylogroups B2 and D, we found in contrast 6 strains belonging to group F, 3 to group A, and only 2 to group B2. We also performed an *in silico* PCR using IPCRESS software to check for the presence of virulence genes. Of the 73 genes evaluated, all 11 sequenced strains harbor at least 30 genes, and 1 strain harbor 48 genes. We are about to receive more NMEC strains, isolated previously and recently, so that we will gather more data on the characteristics of this pathogen and its epidemiology in Brazil.

**Keywords**: neonatal meningitis, *Escherichia coli*, whole genome sequencing, genotyping, phylogenetic typing, virulence

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