

TITLE: Identification and detection of ESBL genes: blaSHV, blaTEM, blaCTXM-1 and blaCTXM-2 and virulence genes: rmpA and wcaG in strains of *Klebsiella* spp. isolated from samples collected at the Human Milk Bank of Vitória da Conquista, Bahia.

AUTHORS: ANDRADE, N. N. N.; SANTANA, L. A. M.; COELHO, L. S.; CORREIA, T. M. L. C.; SOUZA, C. L.; MARQUES, L. M.; OLIVEIRA, M. V.

INSTITUTION: Instituto Multidisciplinar em Saúde, Campus Anísio Teixeira, Universidade Federal da Bahia, Vitória da Conquista, Bahia, Brasil.

ABSTRACT:

Voluntary donors maintain the stock of milked human milk (MHM) from Human Milk Banks (HMB), milk collection can be done at the bank or at home. The hygienic-sanitary conditions used since the collection until the distribution of the MHM by the HMB should follow protocols to avoid the microbiological contamination. Among the bacteria that may contaminate the MHM, *Klebsiella pneumoniae* is a significant cause of neonatal infections, its mechanisms of pathogenicity, virulence and increasing antimicrobial resistance. Therefore, the present study aimed to identify and characterize the pathogenicity and resistance profile of *Klebsiella* spp. in samples from donors from the BLH in Vitória da Conquista. Hands and nipple-areolar region swab, raw human milk (RHM) sample and pasteurized human milk (PHM) from 30 donors enrolled in the HMB were collected. *Klebsiella* strains were isolated in 36.6% (n = 11) of the donors. Of these, 36.6% (n = 4) of the donors presented isolates only in the swab sample, 27,3% (n=3) only in RHM, 18,2% (n=2) with positive results in the swab and RHM samples and 9,1% (n=1) with growth in cutaneous samples, RHM and PHM, obtaining 15 isolates. Through detection of the *pehB* gene by PCR 73.4% (n = 11) were identified as *Klebsiella pneumoniae*. The antibiogram was performed with CLSI 2017 recommendation in all strains: 42.8% (n = 6) presented resistance to Ampicillin and Sulbactam and 35.7% (n = 5) to Amoxicillin and Clavulanic Acid. The isolates were also submitted to the phenotypic detection of ESBL by disk-approximation, 23.1% (n = 3) presented positive results. The qPCR technique was used to detect ESBL genes: SHV-1, TEM, CTXM-1 e CTXM-2 and the virulence genes *rmpA* and *wcaG*. Forty percent (n = 6) presented the *rmpA* gene and 41.6% (n = 5) presented the *wcaG* gene. No strain presented the CTXM-2 gene, 41.6% (n = 5) had the SHV-1 gene, 41.6% (n = 5) the CTXM-1 gene and 53.3% (n = 8) the TEM gene. The isolated PHM sample showed the two virulence genes and three of the four resistance genes. The results of this study point to the possibility of virulent and ESBL-producing *K. pneumoniae* strains reaching the hospital environment through the BLH route from collection to distribution.

Keywords: Human Milk, ESBL, Virulence, real time PCR.

Development Agency: Universidade Federal da Bahia