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# Genomic characterization of hypermucoviscous Carbapenem-resistant *Klebsiella pneumoniae* ST25 isolates from northwest Argentina

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In recent years, an increase in the prevalence hypermucoviscous carbapenem-resistant *Klebsiella pneumoniae* with sequence type 25 (ST25) was detected in hospitals of Tucuman (Northwest Argentina). In this work, a comparative genomic analysis was performed with two *K. pneumoniae* ST25 strains (LABACER 01 and LABACER 27) to characterize the genes associated with virulence and host's colonization. The complete genomes of *K. pneumoniae* LABACER 01 and LABACER 27 were sequenced with the Illumina MiSeq platform (Illumina Inc., San Diego, CA, USA) at INDEAR-BIOCERES (Rosario, Argentina), using a 2\_150 bp read length sequencing protocol. Ribosomal Multilocus Sequence Typing (rMLST) was applied to the *Klebsiella* genomes and 32/51 genes encoding ribosomal protein subunits (rps) were recovered from the species *K. pneumoniae* and queried with the other *Klebsiella* genomes using the BLASTn algorithm. The sequences were concatenated with Mafft software and used for phylogenomic reconstruction with RAxML software. For the reconstruction of the phylogenetic tree, the GTR substitution model and 1000 bootstrap replications were used. Virulence factors associated with *K. pneumoniae* infections were retrieved from the NCBI database and compared across genomes using the BLASTp algorithm. Genomic analysis revealed that *K. pneumoniae* LABACER01 and LABACER27 possess virulence factors found in other strains that have been shown to be hypervirulent, including genes required for enterobactin (entABCDEF) and salmochelin (iroDE) biosynthesis. In both strains, the genes of toxin-antitoxin systems, as well as regulators of the expression of virulence factors and adhesion genes were also detected. Comparative genomics studies performed in this work also showed that the LABACER 01 and LABACER 27 strains possess unique virulence factors when compared to each other, the presence of tamA in the genome of LABACER 01 and not in LABACER 27 could be associated with the ability of the former to colonize the lungs and spread to the blood of infected mice more efficiently. On the other hand, *K. pneumoniae* LABACER 27 possesses the fimbriae genes yadV2, yadV3, and bfpA, associated with the ability of pathogenic *E. coli* strains to colonize abiotic surfaces, as well as to adhere to epithelial cells and even inhibit the phagocytic activity of macrophages. Our genomic study also detected the presence of the rfaH, copA, and aroE genes in the *K. pneumoniae* LABACER 27 genome, were rfaH and aroE are necessary to resist the microbicidal action of the complement system and copA to prevent the bactericidal effect of copper. Studies on the genetic potential of multiresistant *K. pneumoniae* strains as well as their cellular and molecular interactions with the host are of fundamental importance to assess the association of certain virulence factors with the intensity of the inflammatory response. In this sense, this work explored the virulence profile based on genomic and in vivo studies of hypermucoviscous carbapenem-resistant *K. pneumoniae* ST25 strains, expanding the knowledge of the biology of the emerging ST25 clone in Argentina.

**KEYWORDS:** *Klebsiella pneumoniae*; hypermucoviscous; carbapenem resistant; respiratory infection; genomic; sequence type 25.