TITLE: THE INVASIVE AND PERSISTENT *Neisseria meningitidis* MENC CC103 LINEAGE FROM BRAZIL PRESENTS PENICILLIN REDUCED SUSCEPTIBILITY AND IS CHARACTERIZED BY AN ACCESSORY GENE REPERTOIRE.

AUTHORS: FONSECA, E.L.; MARIN, M.A.; ENCINAS, F.; FREITAS, F.; VICENTE, A.C.

INSTITUTION: LABORATÓRIO DE GENÉTICA MOLECULAR DE MICRORGANISMOS, IOC - FIOCRUZ (AV. BRASIL 4365, MANGUINHOS, CEP 21040-360, RIO DE JANEIRO, RJ, BRAZIL).

ABSTRACT

The meningococcal disease is considered one of the major public health concern worldwide. In Brazil, most of meningitis cases occurred in the last decade has been caused by *N. meningitidis* of serogroup C clonal complex ST-103 (MenC CC103), which remains persisting in the country even after the introduction of the MenC conjugate vaccine in 2010. Penicillin is the first line drug for treating meningococcal infection and targets the penicillin-binding protein 2 (PBP2) coded by *penA*. However, the emergence of strains presenting penicillin reduced susceptibility (pen^I) is increasing worldwide, and this phenotype is mainly associated with alterations in PBP2. The aim of this study was to analyze the population structure and accessory genome of MenC CC103 Brazilian strains from a global perspective, and to determine the prevalence of strains presenting the pen^I resistance phenotype and their associated *penA* alleles. Invasive *N. meningitidis* strains were recovered from hospitals in Bahia (2009), Pernambuco (2010–2012), Minas Gerais (2011), and Rio de Janeiro (2012). Complete genome sequencing was performed with the Illumina HiSeq 7500 sequencer for phylogenomic and accessory genome analyses. Several bioinformatics tools were applied for annotation purposes and to assess the resistome, mobilome and pangenome of Brazilian MenC and publicly available genomes. The *penA* allelic diversity was determined using the *Neisseria* MLST scheme. The penicillin MIC was obtained by the E-Test method. The phylogenomic analysis demonstrated that the MenC CC103 corresponded to a lineage responsible by meningitis cases in several Brazilian regions from 2011 to 2015. Comparative genomic analyses revealed the presence of restriction-modification (RMS) and Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) systems. Also, it was identified a genomic island exclusive, so far, to this MenC CC103 lineage, which harboured several genes putatively involved with fitness and virulence. In general, the identified *penA* alleles correlated with the observed penicillin resistance phenotype. Interestingly, the altered *penA14* was found in pen^I invasive MenC cc103 strains spread in Brazil and persisting since 2011. In conclusion, this finding indicates that the biological cost normally imposed by pen^I phenotype can be ameliorated by the particular features identified in this lineage, which represents an additional public health threat.

Keywords: accessory genome, genomic island, *Neisseria meningitidis*, *penA*, penicillin resistance.

Development Agency: Instituto Oswaldo Cruz, CNPq, PNPD/CAPES, Brasil Sem

Miséria (BSM/CAPES), FNS/SVS (25030001256201331).