

Title: GENOTYPIC CHARACTERIZATION BY WHOLE GENOME SEQUENCING OF A GES-5-PRODUCING-*Serratia marcescens*: FIRST REPORT IN BRASIL

Authors: Nodari, C.S.^{1,2}; Soldi, T.^{2,3}; Barth, A.L.^{1,2}.

Institutions: ¹ PPGCF-UFRGS – Programa de Pós-Graduação em Ciências Farmacêuticas da UFRGS (Av. Ipiranga, 2752, Porto Alegre/RS), ² LABRESIS-HCPA – Laboratório de Pesquisa em Resistência Bacteriana do HCPA (Rua Ramiro Barcelos, 2350, Porto Alegre/RS), ³ FACFAR-UFRGS – Faculdade de Farmácia da UFRGS (Av. Ipiranga, 2752, Porto Alegre/RS).

Abstract:

Nowadays carbapenemase production is the main carbapenem resistance mechanism among Enterobacteriaceae. The Guiana Extended-Spectrum (GES) β -lactamase family comprises several Ambler class A enzymes with distinguished hydrolysis profiles. The original GES were classified as ESBL enzymes but amino acid substitutions in the GES-type ESBLs enhanced their activity against carbapenems. The GES-5 is the GES-carbapenemase which hydrolyse imipenem most efficiently. The aim of this work was to characterize genotypically the first GES-5-producing *Serratia marcescens* isolate reported in Brasil. As part of a surveillance study, isolates with reduced susceptibility to carbapenems were submitted to Real Time HRM Multiplex PCR with specific primers to *bla*_{NDM}, *bla*_{KPC}, *bla*_{VIM}, *bla*_{GES}, *bla*_{OXA-48-like} e *bla*_{IMP}. Antimicrobial susceptibility was determined by Etest. Whole genome sequencing (WGS) was performed using the Ion Torrent PGM™ system, with a 400bp-read kit and a 316™ Chip. Library was previously obtained by enzymatic fragmentation. Reads were assembled in contigs and submitted to ResFinder Database. The isolate was obtained from an ascitic fluid recovered in October 2014 from a female patient in a tertiary hospital in Porto Alegre/RS. The Real Time PCR revealed an amplicon with a Temperature of Melting (Tm) similar to the *bla*_{GES} positive control. The isolate presented high-level resistance to carbapenems and polymyxins, remaining susceptible only to fluoroquinolones and tigecyclin. WGS revealed a 5,378,959bp length genome, distributed in 208 contigs (>500bp), with a total GC content of 59%. As expected, the ResFinder results reported the presence of *bla*_{GES-5}, as well as several other β -lactamases, and aminoglycoside resistance determinants. We were also able to observe the presence of genetic resistance markers to sulphonamides and tetracyclins, which we could not determine by phenotypic assays. Interestingly, genetic markers to fluoroquinolones were also pointed out by the ResFinder, regardless the fact that the isolate was susceptible in vitro to these antimicrobials. To the best of our knowledge, this is the first report of a GES-producing *Serratia marcescens* in Brasil. We also demonstrated that NGS platforms can be used as a quick tool to evaluate resistance mechanisms among Enterobacteriaceae.

Keywords: carbapenemases, GES-5, next-generation sequencing.

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