

Quorum quenching activity of *Bacillus cereus* isolate 30b confers antipathogenic effects in *Pseudomonas aeruginosa*

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Abstract

Background: Quorum quenching, the interference of a Quorum sensing (QS) system that contributes to the pathogenesis through triggering the production of various virulence determinants, is among the newly suggested antivirulence strategies.

Purpose: This study aimed at screening of N-Acyl homoserine lactonase activity from local bacterial isolate, and investigating its effect on *Pseudomonas aeruginosa* (*P. aeruginosa*) virulence and biofilm formation.

Materials and methods: Soil bacteria were screened for *aiiA* gene coding for lactonase enzyme by Polymerase Chain reaction and sequencing of *aiiA* gene homologs. Lactonase activity and spectrum were assessed in the cell-free lysate by well diffusion assay using *Agrobacterium tumefaciens* KYC55. A bacterial isolate showing the highest N-acyl-homoserine lactones degradation percentage was identified by gene amplification and sequencing of the 16S rRNA gene and its *aiiA* gene homolog. High performance liquid chromatography was used to confirm N-acyl-homoserine lactone degradation. The effect of cell-free lysate on the biofilm formation ability and cytotoxicity of *P. aeruginosa* PAO1 and *P. aeruginosa* clinical isolates from different clinical sources were assessed by static microtiter plate and viability assay, respectively

Results: Lactonase gene and activity were identified in three *Bacillus* spp. isolates. They showed broad catalytic activities against tested N-acyl-homoserine lactones. However, The lactonase activity in the cell- free lysate of isolate 30b showed the highest significant degradation percentage on all tested signals; N-butanoyl-L-homoserine lactone (71%), N-hexanoyl-L-homoserine lactone (100%), N-decanoyl-homoserine lactone (100%), N-(3-oxohexanoyl)-L-homoserine lactone (37.5%), N-(oxodecanoyl)-L-homoserine lactone (100%), and N-(3-oxododecanoyl)-L-homoserine lactone (100%). Alignment of the amino acid sequences of AiiA protein of isolate 30b showed 96% identity with *Bacillus cereus* (*B. cereus*) homologous lactonases in the GenBank database, and the isolate was designated as *B. cereus* isolate 30b. Cell-free lysate of *B. cereus* isolate 30b reduced biofilm formation significantly in 93% of *P. aeruginosa* isolates. The highest mean percentage of reduction in the biofilm was 86%. Moreover, the viability percentage of human lung carcinoma A549 cells infected by *P. aeruginosa* and treated with cell-free lysate of *B. cereus* isolate 30b increased up to 15%.

Conclusion: The results of this study highlight the potential of lactonases as a promising strategy to combat *Pseudomonas aeruginosa* virulence.

Infection and Drug Resistance 2019, June