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Content

A NOVEL YTNP LACTONASE REDUCES THE EXPRESSION OF *P. AERUGINOSA* MMA83 QUORUM SENSING AND VIRULENCE FACTORS GENE EXPRESSION

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Introduction: Quorum quenching (QQ) is the enzymatic degradation of cell-to-cell signaling molecules. In this study, the potential of the novel YtnP lactonase, the quorum quenching enzyme derived from *S. maltophilia*, to reduce *P. aeruginosa* quorum sensing and virulence factor gene expression was investigated.

Methods: MMA83 culture (adjusted to $1.5x10^5$ CFU/ml) was treated with recombinant YtnP lactonase (final concentration 50 µg/ml) at 37°C for 12 hours under aeration. RNA isolation of the treated and untreated MMA83 culture was performed using the RNeasy Mini Kit (Qiagen, Germany) according to the protocol. Quantitative reverse transcription-polymerase chain reaction (RT-qPCR), was used to analyze the effect of YtnP lactonase on the relative mRNA levels of the LasI/LasR, RhiI/RhiR, and PQS signaling network genes of *P. aeruginosa* MMA83 and virulence factor genes. The *rpsL* was used as an endogenous control to normalize obtained data following the $2^{-\Delta\Delta Ct}$ method.

Results: The QS genes belonging to three QS networks – LasI/LasR, RhiI/RhiR, and PQS of *P. aeruginosa* MMA83 treated with YtnP lactonase were significantly downregulated. The RT -qPCR results show that treatment with YtnP-lactonase decreased the relative mRNA levels of genes involved in the production of elastase (*lasB* approximately 2-fold), alginate (*algK* approximately 2.2-fold), pyocyanin (*phzM* approximately 3.5-fold), pyoverdin (*pvdS* approximately 2-fold), and rhamnolipid (*rhlC* approximately 4-fold). These results suggest that YtnP lactonase exerts an antivirulence effect at the transcription level.

Conclusion: YtnP lactonase, a quorum quenching (QQ) enzyme, has the potential to be used as an innovative enzyme-based antivirulence therapeutic to combat infections caused by *P. aeruginosa*.

Key words: *Stenotrophomonas maltophilia*; lactonases, antivirulence therapy; *Pseudomonas aeruginosa*; quorum sensing

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