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Novel drug targets in cell wall biosynthesis exploited by gene disruption in *Pseudomonas aeruginosa*

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Abstract

© 2017 Elamin et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited. For clinicians, *Pseudomonas aeruginosa* is a nightmare pathogen that is one of the top three causes of opportunistic human infections. Therapy of *P. aeruginosa* infections is complicated due to its natural high intrinsic resistance to antibiotics. Active efflux and decreased uptake of drugs due to cell wall/membrane permeability appear to be important issues in the acquired antibiotic tolerance mechanisms. Bacterial cell wall biosynthesis enzymes have been shown to be essential for pathogenicity of Gram-negative bacteria. However, the role of these targets in virulence has not been identified in *P. aeruginosa*. Here, we report knockout (k.o) mutants of six cell wall biosynthesis targets (*murA*, PA4450; *murD*, PA4414; *murF*, PA4416; *ppiB*, PA1793; *rmlA*, PA5163; *waaA*, PA4988) in *P. aeruginosa* PAO1, and characterized these in order to find out whether these genes and their products contribute to pathogenicity and virulence of *P. aeruginosa*. Except *waaA* k.o, deletion of cell wall biosynthesis targets significantly reduced growth rate in minimal medium compared to the parent strain. The k.o mutants showed exciting changes in cell morphology and colonial architectures. Remarkably, Δ *murF* cells became grossly enlarged. Moreover, the mutants were also attenuated *in vivo* in a mouse infection model except Δ *murF* and Δ *waaA* and proved to be more sensitive to macrophage-mediated killing than the wild-type strain. Interestingly, the deletion of the *murA* gene resulted in loss of virulence activity in mice, and the virulence was restored in a plant model by unknown mechanism. This study demonstrates that cell wall targets contribute significantly to intracellular survival, *in vivo* growth, and pathogenesis of *P. aeruginosa*. In conclusion, these findings establish a link between cell wall targets and virulence of *P. aeruginosa* and thus may lead to development of novel drugs for the treatment of *P. aeruginosa* infection.

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