Characterization of a cytotoxic factor in culture filtrates of *Ralstonia pickettii* RP17 isolated from a water supply pool

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Abstract.

Ralstonia pickettii RP17, isolated from a water supply pool, is a non-pigmented, non-fluorescent and non-fermenting gram-negative bacterium. The effect of cytotoxin produced in culture filtrates of *R. pickettii* RP17 on different mammalian cell lines was investigated. *R. pickettii* RP17, exhibited a dose-dependent cytotoxicity against HeLa, HepG2 cells and Vero cells. Several transposon mutant strains, *R. pickettii* that were deficient in the production of extracellular protease, exhibited significantly attenuated cytotoxicity against mammalian cells. Sequence analysis of the chromosomal DNA flanking the Tn5 insertion indicated Tn5 transposon inserted at an aprX gene which is highly homologous to aprX genes of other closely related aquaculture bacteria *Pseudomonas fluorescens*. Furthermore, heat and EDTA treatments inhibited the cytotoxic activity of *R. pickettii* RP17 culture filtrates towards HeLa cell. These results suggested that *R. pickettii* RP17 derived from aquatic environments can play a pathogenic role in vitro.

Keywords: *Ralstonia pickettii* RP17, culture filtrates, cytotoxin, mammalian cells, aquatic environments