

Construction and characterization of an acapsular mutant of *Pasteurella multocida* strain P-1059 (A:3)

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To further investigate the role of capsule involved in virulence of *Pasteurella multocida* P-1059 (A:3), a *hexB* deleted mutant was constructed by homologous recombination. The DNA replacement was confirmed by PCR, Reverse transcription (RT)-PCR and DNA sequencing. Experiments were conducted to compare the differences of biological characteristics such as capsular structure, capsular polysaccharide content, virulence and serum resistance between the *hexB* deleted mutant of $\Delta hexB$ and wild-type strain P-1059, as well as the complemented strain P-1059C. And the ability of the acapsular mutant $\Delta hexB$ to induced protection against wild-type challenge in chickens. Electron microscopy examination of the $\Delta hexB$ showed the absence of capsular material compared to the P-1059 and P-1059C. The $\Delta hexB$ was sensitive to the bactericidal action of chicken serum, whereas the P-1059 and P-1059C were both resistant. The $\Delta hexB$ was highly attenuated in chickens by intravenously injection, and intramuscular administration of $\Delta hexB$ to chickens stimulated significant protection against P-1059 and the homologous strain X-73(A:1). These results demonstrated that the capsule is a major virulence factor of *Pasteurella multocida* serotype A:3 strains.

Keywords: *Pasteurella multocida*; homologous recombination; knockout; capsule; virulence; vaccine.

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