in equine influenza virus strains and to introduce current isolates into vaccine.

Original papers of this thesis appeared in" Jpn. J. Vet. Res." Vol. 48 : 177-186 and 186-195 (2001) and "Vet. Microbiol." Vol. 20 : 111-119 (2001).

Iron acquisition systems in *Pseudomonas aeruginosa* : Their contribution to bacterial growth and virulence

Hiroyuki Takase

New Product Research Laboratories I
Daiichi Pharmaceutical Co., Ltd.
Tokyo 134-8630, Japan

Pseudomonas aeruginosa is known to possess high-affinity iron acquisition systems mediated by iron chelators produced by the bacterium, generally called siderophores, or operating via heme uptake. This organism produces pyoverdin and pyochelin as siderophores. To investigate the significance of each siderophore for *P.aeruginosa* infection, the author constructed a set of mutants from wild-type strain PAO 1 by allelic exchange, which were deficient in producing one or both of the siderophores. Results of animal experiments using the mutants suggested that both pyoverdin and pyochelin were required for efficient bacterial growth and full expression of virulence in *P.aeruginosa* infections, although pyoverdin might be comparatively more important. However, the siderophores were not always required for the infections. Considering results of in vitro experiments, it was thought that iron acquisition via heme uptake might also play an important role in *P.aeruginosa* infections.

High-affinity iron acquisition systems of gram-negative bacteria involve specific receptors in the outer membrane. The transport of iron-siderophore complexes and heme bound to the receptors into the periplasm is generally thought to be dependent on the function of the cytoplasmic protein, TonB. To clarify the contribution of the TonB protein to high-affinity iron acquisition in *P.aeruginosa,* the author constructed *tonB*-inactivated mutants from strain PAO 1 and its siderophore-deficient derivative. Results of in vitro growth assays of them indicated that the TonB protein was essential for iron acquisition mediated by pyoverdin and pyochelin and via heme uptake in *P.aeruginosa.* In addition, results of animal experiments suggested that the TonB-dependent iron acquisition might be essential for *P.aeruginosa* to infect the animal host.

Original papers of this thesis appeared in" Infect. Immun." Vol. 68 : 1834-1839 and 4498-4504 (2000).