

RAPID IDENTIFICATION AND VIRULENCE CHARACTERIZATION OF *BACILLUS ANTHRACIS* USING PYROSEQUENCING™ TECHNOLOGY

A. Asp², H. Kling¹, S. Hjalmarsson¹, A. Alderborn², C. Fock¹, I. Muldin², U. Eriksson³, F. Elgh³, L. Engstrand^{1,4}

¹Dept. of Medical Sciences, Clinical Bacteriology, Uppsala University, Uppsala, Sweden

²Pyrosequencing AB, Uppsala, Sweden

³Swedish Defence Research Agency, Umeå, Sweden

⁴Swedish Institute for Infectious Disease Control, Stockholm, Sweden

The bacterium *Bacillus anthracis* is a major threat in biological warfare. Inhalation of spores causes inhalation anthrax, a serious infection leading to death in untreated cases. Rapid, secure identification and virulence characterization of the pathogen is therefore crucial for diagnosis determination and choice of an optimal treatment regimen.

Sequence analysis of the 16S rRNA gene, commonly used for bacterial typing, cannot be used in this situation since *B. anthracis* and *B. cereus* have identical sequences. A DNA sequence called Ba813 has been used instead as a specific chromosomal marker for *B. anthracis*, making it possible to distinguish this strain from other closely related soil-borne *Bacillus* species. Moreover, virulent strains of *B. anthracis* harbour two plasmids, pXO1, containing genes for production of anthrax toxin and pXO2, encoding products necessary for encapsulation and spore formation. We employed PCR together with Pyrosequencing™ technology for species identity confirmation and virulence characterization of 10 *B. anthracis* isolates.