THE ROLE OF SPORE COAT ON SPORE GERMINATION

CHEUNG SAU HA

MASTER OF PHILOSOPHY
CITY UNIVERSITY OF HONG KONG
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City University of Hong Kong
香港城市大學

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胞子套對胞子發萌的角色

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Cheung Sau Ha
張秀霞

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ABSTRACT

Bacterial spores exhibit extreme metabolic dormancy. They are capable to survive under a range of harsh conditions which would rapidly kill vegetative cells. Despite this extreme dormancy, spores retain an alert mechanism allowing them to respond to a favourable environment by instant germination. This response consists of probably a complex biophysical followed by biochemical events which result in the breakdown of the dormant state and resume vegetative growth. Germination has been found to be a process of irreversible degradation of chemical reactions, i.e. activation, initiation and outgrowth. Once spores commit to germinate, the germinant-mediated response associated with initiation constitutes the trigger reaction and thus, would irreversibly lose their unique spore properties. However, spore germination can be affected by medium in which the spores were formed. In this study, we hypothesize that disruption of the coat proteins resulted in poorly germinating or completely dormant form of spores even in the presence of germinant. By comparing the germination profile of defined *Bacillus subtilis* spores prepared in different types of nutrient-limiting media, significant differences in germination rate were observed amongst three types of nutrient-depleted spore. Nitrogen-depleted spores, in general, germinated very well. About 83% of these spores changed from phase dark to phase-bright under microscope while
carbon-depleted spores germinated slowly (25%) and sulphur-depleted spores were totally dormant. Electron microscopic analysis revealed that the coat layer of the later type of spores was defected while the former was intact. Ridge formation and small bumps were observed on the surface of wild type spores and nitrogen-depleted spores while absent in sulphur-depleted spores and decoated spores. This observation was in agreement to data derived from Fourier-Transform Infrared (FTIR) analysis; in which sulphur-depleted spores had less amino and amide components while nitrogen-depleted spores didn’t. To test whether spore coat layer were really required for spore germination, the coat layer of nitrogen-depleted spores of *B. subtilis* was either physically or chemically stripped off. In both cases, it was noted that spore germination was significantly reduced. On the other hand, when spore coat was embedded with low concentration of alkaline glutaraldehyde, a strong cross-linking agent which can fix proteins in the spore coat, blockage of spore germination was observed. These changes were in alliance with the differences of coat protein of nutrient depleted spores and those of coat defected spores of *B. subtilis*. All these results, therefore, prove that some proteins in the spore coat, which is the foremost accessible structure of a spore, are likely the triggering apparatus responsible for swift response of spore to germinant in the germination process.
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