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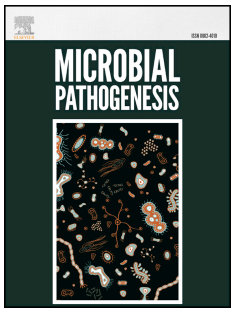

Novel motB as a potential predictive tool for identification of *B. cereus*, *B. thuringiensis* and differentiation from other *Bacillus* species by triplex real-time PCR

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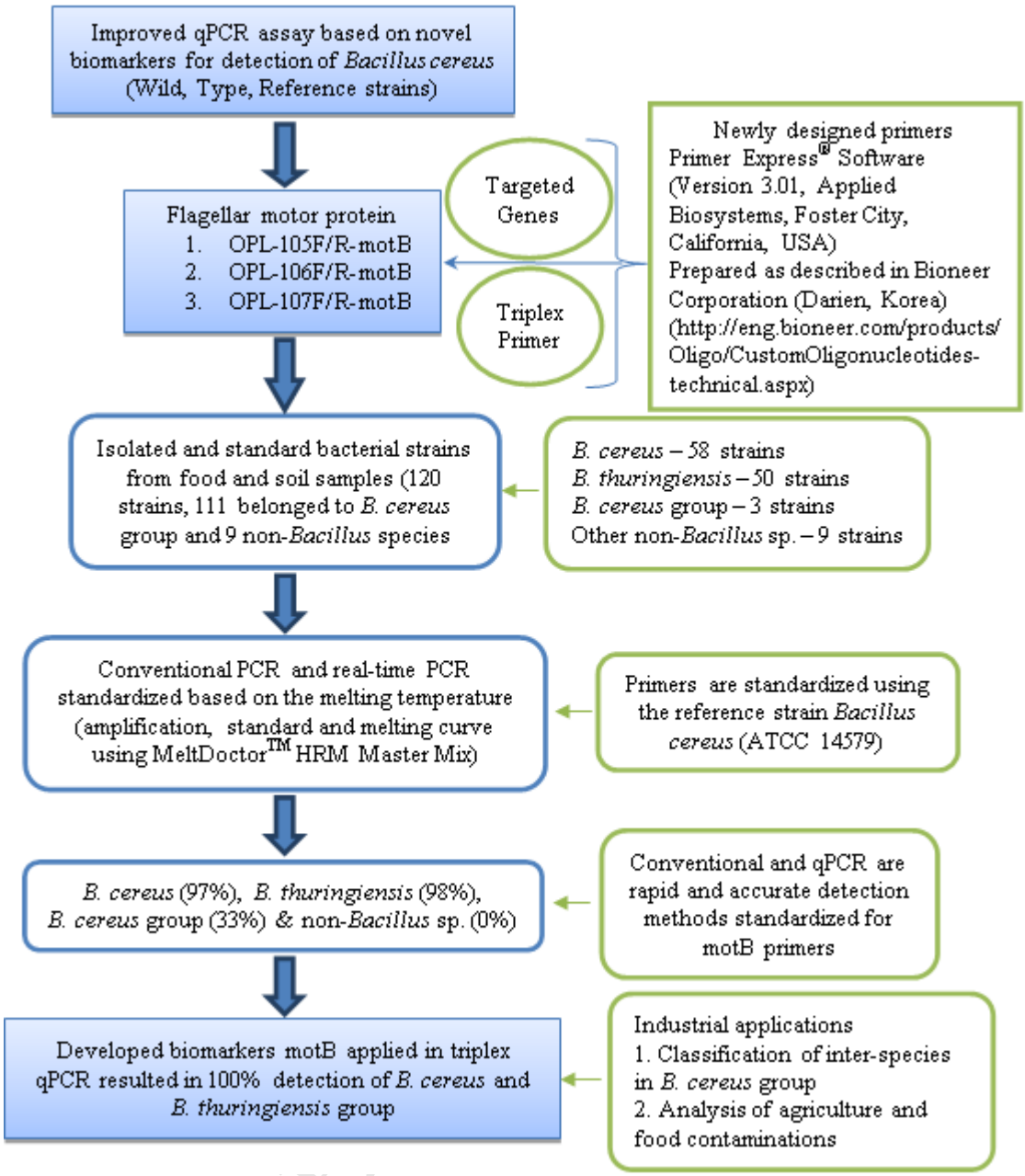
Improved qPCR assay based on novel biomarkers for detection of *Bacillus cereus* (Wild, Type, Reference strains)
Flagellar motor protein
1. OPL-105F/R-motB
2. OPL-106F/R-motB
3. OPL-107F/R-motB
Targeted Genes
Triplex Primer
Newly designed primers Primer Express® Software (Version 3.01, Applied Biosystems, Foster City, California, USA) Prepared as described in Bioneer Corporation (Darien, Korea) (http://eng.bioneer.com/products/Oligo/CustomOligonucleotides-technical.aspx)
Isolated and standard bacterial strains from food and soil samples (120 strains, 111 belonged to *B. cereus* group and 9 non-*Bacillus* species
B. cereus – 58 strains
B. thuringiensis – 50 strains
B. cereus group – 3 strains
Other non-*Bacillus* sp. – 9 strains
Conventional PCR and real-time PCR standardized based on the melting temperature (amplification, standard and melting curve using MeltDoctor™ HRM Master Mix)
Primers are standardized using the reference strain *Bacillus cereus* (ATCC 14579)
B. cereus (97%), *B. thuringiensis* (98%), *B. cereus* group (33%) & non-*Bacillus* sp. (0%)
Conventional and qPCR are rapid and accurate detection methods standardized for motB primers
Developed biomarkers motB applied in triplex qPCR resulted in 100% detection of *B. cereus* and *B. thuringiensis* group
Industrial applications
1. Classification of inter-species in *B. cereus* group
2. Analysis of agriculture and food contaminations

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