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Whole-cell proteome analysis of *T. thermophilus* HB8 by using nano-LC and Q-TOF

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Thermus thermophilus HB8, an extremely thermophilic bacterium, is a model microorganism for basic bacteriology and industrial application due to its thermophilic enzymes, simple cultivation, easy gene manipulation and relatively small number of orfs encoded in the genome. *T. thermophilus* HB8 encodes 2,239 orfs including about 30% unknown function and conserved hypothetical proteins in a chromosome and two plasmids (pTT27 and pTT8). The study of *T. thermophilus* physiology using proteomics enables more predictive understanding of biological phenomena that response to environmental stimuli and different culture conditions.

We carried out nano-scale liquid chromatography (nLC) combined tandem mass analysis to investigate protein expressions in *Thermus thermophilus* HB8 under whole-cell level. The crude extracts were prepared from *T. thermophilus* HB8 cells cultivated in complex media or minimal media at its early stationary phase. To decrease complexity of proteins and increase identification number of peptide and protein by MS/MS analysis, the whole cell proteins were further separated by gel filtration, ion-exchange or hydrophobic chromatography prior to fragmentation by trypsin. In this study, 1,394 different proteins with 17,128 unique peptides were identified from 211,132 MS/MS results derived from 169 nLC-MS/MS analyses. The number of the identified proteins reached ~62.2% of total ORFs in the genome of *T. thermophilus* HB8. This result provides lots of information about the expressed proteins in *T. thermophilus* HB8 such as expression of hypothetical proteins, protein-protein interaction, N-terminal peptide sequence and post-translational modification of a protein.