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Review of doctoral thesis of
Mashzhan Akzhigit Sembayuly
for the Doctor of Philosophy (Ph.D) degree

“Metagenomic analysis of Kazakhstan thermophilic bacteria for obtaining promising hydrolytic enzymes”

The dissertation work of Mr. Mashzhan Akzhigit Sembayuly focuses on the microbial diversity of extremophiles in geothermal springs in Kazakhstan using cultivation-independent (metagenomics based) and cultivation-dependent approaches with the aim to discover novel biotechnologically useful enzymes and organisms. Geothermal environments in Kazakhstan have previously not been subjected to microbial investigations and thus represent a vast unexplored resource for discovery of novel thermophilic microbes with industrial and biotechnological potential. Mr. Mashzhan Akzhigit Sembayuly's dissertation consists of an extensive introduction to the subject of extremophiles and thermophiles and an extensive results and methods part from 11 scientific publications, including 3 articles in domestic periodicals, one book chapter in *Microbial Communities and their Interactions in the Extreme Environment* (Springer book), one published research article in *Current Microbiology* as well as one paper which currently is in the second round of review in *International Journal of Systematic and Evolutionary Microbiology*. Mr. Akzhigit is the first co-author in all these publications showing impressively his research potential and network of collaborators.

During the last couple of decades, agro-industrial and other biotechnology- and enzyme-based industries have increased their use of robust enzymes (thermozymes) from extremophiles in large-scale processes. Thermozymes have superior properties compared with traditional enzymes from mesophilic microorganisms as they are heat-tolerant and also tolerate harsh conditions like extreme pH values and presence of organic solvents, which are often associated with large-scale and industrial processes. Mr. Akzhigit has mainly focused on extremophiles with hydrolytic enzyme production e.g. proteases, cellulases, amylases and lipases, all highly industrially relevant enzymes.

Mr. Akzhigit's in-depth diversity analyses of the microbial communities in Kazakh hot springs, revealed a dominance of extremophiles belonging to the *Bacilli* class. Some of these organisms, representing *Geobacillus* spp. were, furthermore, isolated and identified, and found to produce various hydrolytic enzymes active from 55 to 75°C. These isolates represent a high biotechnological potential. Some of the isolates appear to represent novel *Geobacillus* species and are therefore also highly interesting from a taxonomic viewpoint as they enhance the knowledge of the diversity, biogeographical distribution pattern and possibly an allopatric evolution of this microbial group. Another isolate, belonging to genus *Polycladomyces* within family *Thermoactinomycetaceae*, appeared to be particularly interesting and was described as a novel species, *Polycladomyces zarkentensis* sp. nov., and represents the third characterized species of this underexplored but widely distributed bacterial genus. This isolate is of particular interest as it grows fast and produces four classes of hydrolytic enzymes at elevated temperatures, particularly a thermoactive cellulase with optimal production/activity at 70°C. A polyphasic taxonomic analysis including 16S rRNA gene sequencing, whole genome sequence analysis, fermentation pattern, enzyme profiling, and cellular fatty acid profile analyses, firmly placed the isolate into a novel and distinct *Polycladomyces* species, representing a significant contribution to developing the taxonomic framework for this genus.

Mr. Akzhigit demonstrates an excellent and deep understanding of the microbiological and molecular methods used in his work. He furthermore shows a very good understanding of the interpretation of the results in the context of microbial diversity and biotechnological potential. It is the overall impression that his work is at a high scientific quality, and that Mr. Akzhigit works quite independently. In my opinion the thesis fulfills all requirements for obtaining a PhD degree and is ready to be defended in front of a respective committee.

Sincerely yours,


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