ABSTRACT BOOK

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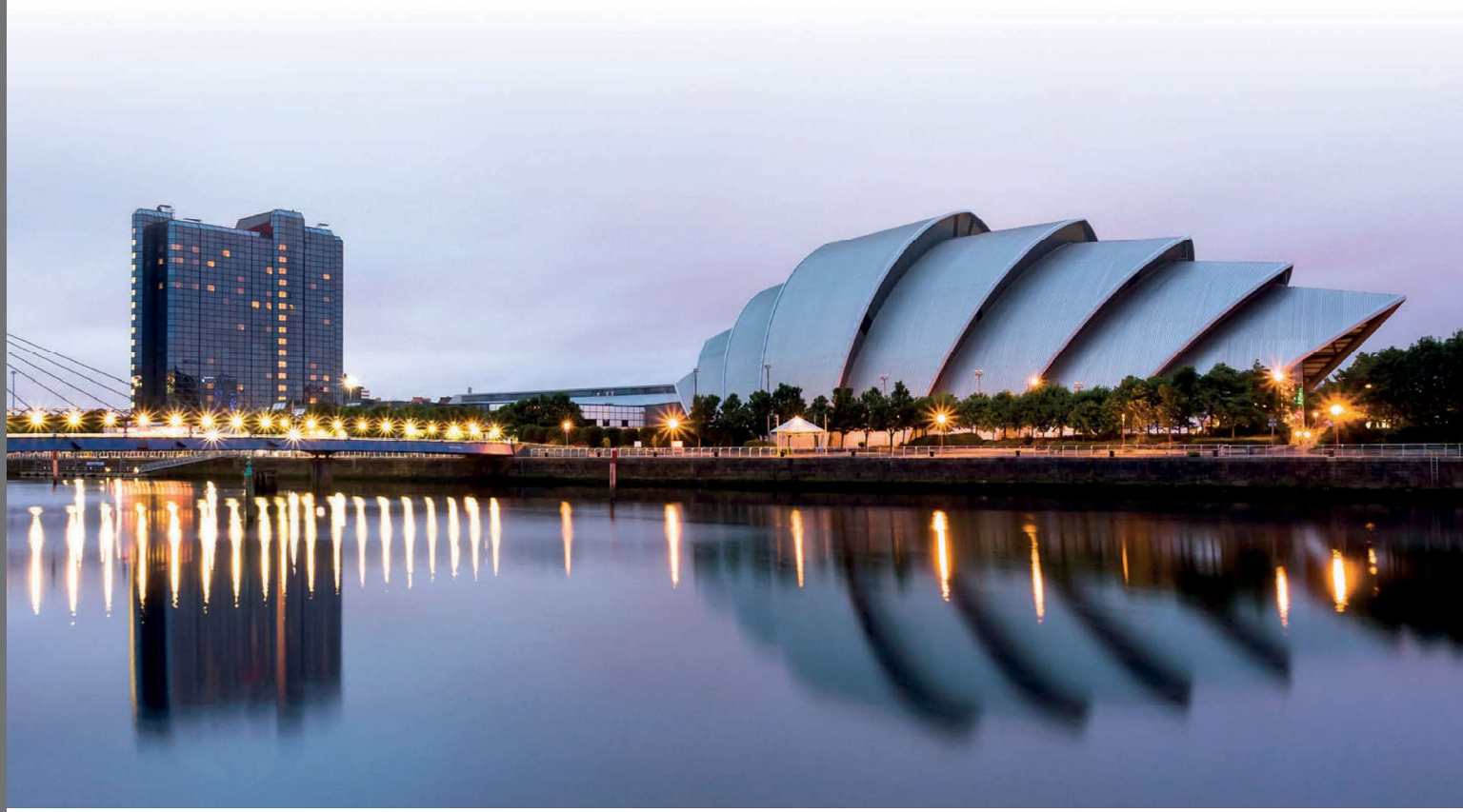

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PT278 Identification of novel amyloid-forming proteins RopA and RopB in the root nodule bacterium Rhizobium leguminosarum

Anastasiia Kosolapova[1;2], Kirill Antonets[1;2], Mikhail Belousov[1;2], Maria Belousova[1], Anna Lykholay[2], Oksana Shtark[1], Ekaterina Vasileva[1;2], Kirill Volkov[2], Anton Nizhnikov[1;2]

[1]*All-Russia Research Institute for Agricultural Microbiology, St. Petersburg, Pushkin, Russian Federation*
[2]*St. Petersburg State University, St. Petersburg, Russian Federation*

Background: Amyloids represent non-soluble protein aggregates formed by unbranching fibrils characterized by cross-β structure. The amyloid formation is associated with development of more than 40 fatal diseases including Alzheimer's disease. Nevertheless, investigations of last decades demonstrated that amyloids can perform various physiological functions. The majority of functional amyloids of prokaryotes were identified within *Gammaproteobacteria* species including *Escherichia coli*. However, no functional amyloids have been identified within *Alphaproteobacteria* species yet.

Objectives: We aimed to investigate novel functional amyloids of *Rhizobium leguminosarum* (class *Alphaproteobacteria*) – agriculturally important symbiont of legumes.

Methods: To identify potential amyloidogenic proteins, we used previously developed Proteomic Screening and Identification of Amyloids (PSIA) approach. Analysis of amyloid properties of proteins was performed in Curli-Dependent Amyloid Generator (C-DAG) system, which allows to export a protein of interest to the surface of *E. coli* cells.

Results: We identified 54 proteins in detergent-resistant fraction of *Rhizobium leguminosarum* using PSIA approach. For further analysis, we chose two outer membrane proteins involved in nodulation - RopA porin, identified with the highest mass-spectroscopy score, and outer membrane protein RopB. We demonstrated that cells exporting RopA and RopB proteins exhibit typical for amyloids apple-green birefringence in polarized light upon Congo Red dye binding. Also, RopA and RopB form unbranched fibrils on the cell surface. Moreover, RopA and RopB proteins fused with YFP aggregate in yeast cells. Taking together, we may conclude that RopA and RopB are able to form amyloid fibrils.

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