## Characterization of Polylactic Acid Degrading Bacterium strain N-3 from Antarctic Soil

Kenta Sato<sup>1</sup>, S. A. Ahmad<sup>2</sup> and A. Zulkharnain<sup>1</sup>

<sup>1</sup>Department of Bioscience and Engineering, College of Systems Engineering and Science, Shibaura Institute of Technology, 307 Fukasaku, Minuma-ku, Saitama, 337-8570, Japan

<sup>2</sup>Department of Biochemistry, Faculty of Biotechnology and Biomolecular Sciences, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

Plastic is widely used in our daily life. However, it is difficult to be degraded in the natural environment and remains persistent for an extended period of time. In this environmental problem, ecologically friendly bioplastics have been attracting a lot of attention as these categories of plastics can be degraded by microorganisms. To date, several strains of bacteria including psychrophilic have been reported to possess the ability to degrade different types of plastics1). Polylactic acid (PLA) is a type of bioplastic that is synthesized from biological resources and is biodegradable. It is used for prosthetic devices and food packaging due to its biologically safe characteristic. In addition, its strength and functionality are being studied to improve its usability as a plastic. Previously, we reported on Massilia (Naxibacter) sp. N-3 strain isolated from Antarctic soil sample, collected from Signy Island, with confirmed ability to degrade PLA. Strain N-3 degraded PLA granules for four weeks. The objective of this study was to investigate the biochemical, microbiological, and genetic properties of Massilia sp. N-3 strain, in order to characterize the strain and elucidate the mechanism of PLA degradation.

Comparison of growth at different incubation temperatures revealed that strain N-3 was not a psychrophilic bacterium but a psychrotolerant. The genomic DNA of this bacterium was extracted and whole genome sequencing was performed. Sequencing was performed on Illumina Hiseq and PacBio RS II platforms, and assembly results revealed the sequence to have a length of 5.58 Mbp, an average coverage of 136.05, and a GC content of 65.44%. The gene annotation results that it had 5187 genes (including RNA genes) and identified eight genes, including PHB depolymerase and hydrolytic enzymes, as PLA-degrading genes. In addition, three sequences were annotated as unknown genes belonging to alfa/beta hydrolases. Three and seven genes were identified as esterases and proteases, respectively, which are known enzymes that degrade PLA. Besides, genes for enzymes that assimilate lactic acid, a metabolite of PLA, were identified. Strain N-3 was found to possess dehydrogenase genes that metabolize L- and D-body lactic acid, respectively. These results indicated that strain N-3 has complete genes to metabolizes PLA.

In the future, we will use RT-PCR to confirm whether this enzymatic expression during PLA degradation, and conduct functional analyses of these genes using *E. coli* as host.

## References

## References

1)Atanasova, N.; Stoitsova, S.; Paunova-Krasteva, T.; Kambourova, M. Plastic Degradation by Extremophilic Bacteria. *Int. J. Mol. Sci.* **2021**, 22, 5610. DOI: 10.3390/ijms22115610