

## Effect of caffeine on the stress resistance of *Lactobacillus paracasei*

Hiroaki Sato<sup>1</sup>, Toru Mizuki<sup>2</sup>, Takeshi Miura<sup>1</sup>

<sup>1</sup>Graduate School of Science, Toyo University, Itakura, Gunma, 374-0193 Japan

<sup>2</sup>Bio-Nano Electronics Research Centre, Toyo University, Kawagoe, Saitama, 350-8585 Japan

Caffeine, is known as a component of coffee, has antibacterial activity against not only pathogenic bacteria but also against beneficial bacteria those are used for fermentation, food processing and other industrial use. We discovered *Lactobacillus paracasei* YSAK1 and YKP4 strains, those are caffeine-resistant bacteria. Environmental stresses, such as pH, temperature, pressure and chemical substance have various effect on the viability, surface form and composition of membrane of lactic acid bacteria. The objective of this study is to find out the difference of the effect of caffeine on wild type and caffeine resistant strains of *L. paracasei*.

We used two caffeine-resistant bacteria and a wild type strain, that is caffeine sensitive, as a control to investigated the effect of caffeine on lactic acid bacteria. The bacteria were cultured with liquid medium with/without caffeine. Bacterial viability was estimated by measurement and calculation of colony forming unit. We observed the morphological changes on the surface of the bacterial membrane with a scanning electron microscope (SEM). We analyzed the difference of membrane composition of three strains by Sherlock Microbial Identification System.

Depend on the increase of the caffeine concentration contained in the culture medium, the wild type strain showed viability decrease, cell elongation, and perforation of the cell surface, but the caffeine resistant strains showed a high viability and normal bacterial membrane. It has been confirmed that the ratio of saturated fatty acids in the wild type strain and unsaturated fatty acids in caffeine resistant strains were increased, respectively, due to caffeine treatment of cell membranes.

YKP4 strain has a high resistance to caffeine and a high content of unsaturated fatty acids compared to YSAK1 strain. Furthermore, it has been observed that YSAK1 strain elongates in morphology due to culture in the presence of high-concentration caffeine.

These results indicate that there is a difference in the effect of the wild type strain and the resistant strains on caffeine, and even in resistant strains, there is a difference between YSAK1 strain and YKP4 strain.