Isolation, characterization and control of Shiga toxin-producing Escherichia coli by using lytic bacteriophages

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## 論文題名 : Isolation, characterization and control of Shiga toxin-producing *Escherichia coli* by using lytic bacteriophages

(志賀毒素産生性大腸菌分離株の性質と溶菌ファージによる制御)

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論文内容の要旨

Shiga toxin-producing *E. coli* are lethal pathogens responsible for frequent outbreaks of gastroenteritis in humans with symptoms ranging from watery diarrhea to hemorrhagic colitis, and even more complicate illness such as hemolytic uremic syndrome. Most of STEC infections in human are attributed to consumption of foods contaminated with these pathogens including raw or undercooked meat fresh produce, and juice. Owing to eminent threats of STEC to human health, methods for elimination of STEC contamination in foods. Lytic bacteriophages (phages) are natural antimicrobials, abundant in the environment, and non-toxic for humans and have successfully been demonstrated as promising agents for decontamination of important foodborne bacteria.

The focus of this study was to investigate the STEC contamination in raw meat samples, which was followed by the isolation and application of lytic phages to control STEC O157:H7. The data obtained from current study showed that meat samples analyzed, except raw chicken meat, were contaminated with STEC at the rate of 4.4%. Even though most STEC strains isolated from raw meat had virulence gene composition less complicate compared to that of clinical STEC, several of them also contained virulence factors very similar to those found in clinical STEC suggesting that they are possibly pathogenic for humans. Seven lytic phages against STEC strains including serotype O157:H7 were also successfully isolated from bovine intestine samples. The characterization results revealed that three phages (PE37, PE96, and PE110) that belong to the *Myoviridae* family have the attributors of a phage potentially used as bactericide for STEC. The treatment of STEC O157:H7 at both in broth culture and in raw beef meat samples with the isolated phages showed significant reductions of bacterial viable counts compared to non-treatment. For instance, the co-incubation of phage PE37 with STEC O157:H7 in LB broth at 37°C after 4 h caused a reduction in viable counts by 5.15 log CFU/ml or phage PE96 reduced cell concentrations of STEC O157:H7 by 3.89 log CFU/ml when they were incubated in LB at 25°C after 4 h. In the raw beef pieces, the treatment of samples experimentally contaminated with STEC O157:H7 with phage PE37 at 25°C and 8°C after 24 h induced the bacterial reductions of 2.33 and 0.93 log CFU/piece, respectively, whereas in almost similar trials, phage PE96 reduced viable counts of STEC O157:H7 by 0.92 and 1.5 log CFU/piece after 24 h at 25°C and 8°C, respectively.

The findings of this study showed the presence of some STEC strains potentially pathogenic for humans in raw meat samples and demonstrated the possible applications of lytic phages to control STEC O157:H7 in raw beef. The use of these lytic phages alone and in combination with other antimicrobial agents or food additives for controlling pathogenic STEC strains present in various types of foods could also be a possible trend in the future.