

E303 Purification and Characterization of Peptidase Having Fibrinolytic Activity from *Bacillus atrophaens*

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We have isolated a extracellular serine protease having fibrinolytic activity from *Bacillus atrophaens* screened from Doen-Jang, a traditional soybean fermented food in Korea. The enzyme was purified by two successive chromatographic techniques, DEAE-cellulose followed by the gel filtration. Maximal activity of the enzyme occurs at pH 9.0 and 55 to 60 °C. The isolated protein with an apparent size of 42 kDa on SDS-PAGE and N-terminal 18 amino acid sequence appeared to be identical to that of previously described peptidase from *Bacillus*. The of enzyme was inhibited by PMSF and SDS, but not leupeptin and EDTA.

E304 Chemical Modification of a Levanheptaose-Producing Levanase and Effect of Levanheptaose on Intestinal Microflora

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Levanase from *Streptomyces sp.* is inactivated by several modifying chemicals such as, 2-Hydroxy-5-Nitrobenzyl Bromide, N-Bromosuccinimide, Diethyl pyrocarbonate and SH-group reagents. The inhibition by 2-Hydroxy-5-Nitrobenzyl Bromide and N-Bromosuccinimide were prevented by 1% levan. These results indicate that histidine and cysteine residue are essential for the enzyme activity and tryptophane may be in the active site of levanase.

And, we investigated the effect of levanheptaose on principal intestinal microflora in two experiments. In the *in vivo* experiments, intestinal microflora were cultured on a medium containing levanheptaose. *Bifidobacterium adolescentis* and *Lactobacillus acidophilus* grew effectively, whereas *Clostridium perfringens*, *Bacillus fragilis* and *Staphylococcus aureus* did not. Therefore, levanheptaose seem to promote selectively the growth of *B. adolescentis*. In the *in vivo* experiments, we examined the effects of levanheptaose on intestinal microflora, pH, and butyrate concentration in rats. Apparently, fecal *Bifidobacteria* was higher, whereas total aerobes were lower in rats fed levanheptaose diets compared with those fed the control diets. We concluded that dietary incorporation of levanheptaose by providing butyrate, lowering pH, and increasing *Bifidobacteria* may be beneficial in improving gastrointestinal health.