

(学位第3号様式)

学 位 論 文 要 旨	
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題 目	Studies on Thermotolerant Acetic Acid Bacteria from Sri Lanka: Taxonomic Characterization and Analyses of Membrane-bound Enzyme Activities and Pellicle Polysaccharides (スリランカからの耐熱性酢酸菌に関する研究: 分類学的特徴及び膜結合型酵素活性と菌膜多糖の解析)
<p>Acetic acid bacteria (AAB) are of obligate aerobic Gram-negative bacteria with the ability to oxidize ethanol to acetic acid. Among them, <i>Acetobacter</i> and <i>Gluconacetobacter</i> are used in industrial vinegar production. Since heat is generating during fermentation processes, a large cooling system is required for maintaining the optimum temperature, which increases the production cost, especially in tropic and sub-tropic area. Thus, development of thermotolerant strains would be beneficial. In this thesis, isolation, characterization, and analysis of membrane bound enzymes and pellicle polysaccharides of thermotolerant acetic acid bacteria from Sri Lankan coconut water vinegar has been done.</p> <p>From the pellicle formed on the top of brewing coconut water vinegar in Sri Lanka, three <i>Acetobacter</i> strains (SL13E-2, SL13E-3 and SL13E-4) which grow at 42°C and four <i>Gluconobacter</i> strains (SL13-5, SL13-6, SL13-7 and SL13-8) which grow at 37°C were identified as <i>Acetobacter pasteurianus</i> and <i>Gluconobacter frateurii</i> respectively. The three <i>Acetobacter</i> strains could tolerate up to 8% and 3% ethanol concentration at 41°C and 42°C, respectively. Acetic acid production by the isolated <i>Acetobacter</i> strains was examined: all three strains gave 4% acetic acid at 37°C and 6% initial ethanol, and at 40°C provided 2.5% acetic acid with 4% initial ethanol. Compared with the two other strains, SL13E-4 showed both slower growth and slower acetic acid production rate. As well as thermotolerant SKU1108 strain, activities of ADH and ALDH of SL13E-2 and SL13E-4 were more stable than those of the mesophilic strain. The isolated strains were used to produce coconut water vinegar at higher temperatures than typically used for vinegar production.</p> <p><i>Acetobacter</i> species are well known to have an ability to grow floating on the surface of the medium by producing pellicle, which consists of cells and a self-produced matrix of cell-attached polysaccharide. Thus, characterization of pellicle polysaccharides of <i>A. pasteurianus</i> SL13E-2, SL13E-3, and SL13E-4 from Sri Lankan coconut vinegar and <i>A. pasteurianus</i> SKU1108, which was originally isolated from Thailand, was done. The monosaccharide composition of pellicle polysaccharides of <i>Acetobacter pasteurianus</i> strains found to be varied among strains. The pellicle polysaccharide of SL13E-2 composed of rhamnose and glucose in the ratio of 1:8 and that of SL13E-4 and mesophilic <i>A. pasteurianus</i> NBRC3191 consisted of rhamnose, glucose and xylose in the ratio of 1:5:2 and 1:4:2 respectively. On the other hand, the pellicle polysaccharides of SL13E-3 and SKU1108 strains were composed of rhamnose, glucose and galactose in the ratio of 2:2:1 and 1:5:2.5 respectively. The pellicle formation of thermotolerant SL13E-2, SL13E-3 and SL13E-4 were significantly induced by the addition of ethanol, while a poor induction was observed in SKU1108 strain. The size and sugar composition of the polysaccharides obtained from the cells induced by ethanol were unchanged from those from the uninduced cells, indicating that number of molecules of the polysaccharides increased and not changed the individual molecule of polysaccharide. On the other hand, addition of sugar sources, such as glucose, sucrose and fructose, showed a slight induction of pellicle formation in SKU1108 strain, especially at 40°C.</p>	