

Pawinee Wongtubtim 2012: Production of  $\alpha$ -Amylase and Probiotic Bacteria by Solid State Fermentation using Cassava Chips as Raw Material. Master of Science (Microbiology), Major Field: Microbiology, Department of Microbiology.

Thesis Advisor: Associate Professor Vichien Kitpreechavanich, Dr.Eng. 118 pages.

Production of probiotic and  $\alpha$ -amylase were investigated by solid state fermentation using cassava chip as raw material. Therefore, *Bacillus* strains as probiotics and high  $\alpha$ -amylase producing *Rhizopus* strains with raw starch digestion were selected. Among one hundred-six strains of *Bacillus* spp., 10 strains exhibited the ability to inhibit *Staphylococcus aureus*, *Escherichia coli*, *Enterococcus aerogenes* and *Salmonella* Typhimurium. The strains KMS 2.1-1, KMS 2.2-1 and FB 11 could be tolerant at pH 2 with 63.4, 62.7 and 58.4% survival rate, respectively and to 0.3% bile salt with 52.4, 85.4, 67.9 % survival rate. These strains were found to be able form biofilms. Strains of KMS 2.1-1 and FB 11 showed strong  $\beta$ -hemolysis activity on sheep blood agar, whereas the strain of KMS 2.2-1 showed weak  $\beta$ -hemolysis activity but no toxic signs and mortality in acute oral toxicity test to mice. Selection of *Rhizopus* strains on  $\alpha$ -amylase production ability with raw cassava starch digestion was performed using cassava chip as substrate by solid state fermentation. Strain of *R. oryzae* TISTR 3514 was the strain that produced high  $\alpha$ -amylase production and raw cassava starch digestion. Therefore, *R. oryzae* TISTR 3514 were selected for co-culture with *Bacillus* KMS 2.2-1 by solid state fermentation. At 6 days of fermentation, the number of cells was  $2.1 \times 10^{10}$  cell/g dry substrate and  $\alpha$ -amylase activity had 43 unit/g dry substrate. Whereas the monoculture of *Bacillus* KMS 2.2-1, the number of cells and  $\alpha$ -amylase activity had  $4 \times 10^{10}$  cell/g dry substrate with 50 unit/g dry substrate, respectively. The results indicated the antagonist of *Bacillus* KMS 2.2-1 to *R. oryzae* TISTR 3514. In pararell, *R. oryzae* TISTR 3514 and *Bacillus* sp. A5 previous reported as probiotic were cocultured by solid state fermentation, at 6 days of fermentation, the number of *Bacillus* cells was  $6 \times 10^9$  cell/g dry substrate and  $\alpha$ -amylase activity was 16 unit/g dry substrate with raw starch digestion of 1.18 unit/g dry substrate.

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Student's signature

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Thesis Advisor's signature