Ubonphan Laungubon 2007: Screening of Thermotolerant Methylotrophic Bacteria Capable of Formaldehyde Fixation. Master of Science (Microbiology), Major Field: Microbiology, Department of Microbiology. Thesis Advisor: Mrs. Nantana Srisuk, Ph.D. 165 pages.

Seventy-six isolates of methylotrophic bacteria were isolated on methanol agar medium from 20 natural samples but only 24 isolates exhibited growth in methanol broth at 45°C. Among these, 19 isolates were found to be thermotolerant methylotrophic bacteria capable of methanol utilization for growth. Morphological study of bacterial cells showed that 17 isolates possessed rod-shaped cells whereas the other 2 isolates were cocci, gram positive and non-endospore forming bacteria. Among the rod -shaped cells group, 6 isolates were found to be gram positive and endospore former whereas 5 isolates were also gram positive but non-endospore former. The other 2 isolates were gram variable and endospore forming bacteria. The last 4 isolates of this group were gram negative of which 3 isolates were endospore former and a single isolate was non-endospore former. Screening for thermotolerant methylotrophic bacteria capable of formaldehyde fixation at 45°C was performed by determining the minimum inhibitory concentration (MIC) of formaldehyde to the bacterial growth in methanol broth. Results indicated that the methylotrophic bacteria isolate SB4, TE1 and TB2 possessed the highest MIC as 2.5, 2.4 and 1.9 mM, respectively. MICs of formaldehyde to the respective bacterial growth under the condition that formaldehyde was the sole carbon and energy source were found to be 0.8, 0.7 and 0.5 mM, respectively. Capability of the bacterial isolates SB4, TE1 and TB2 were therefore assessed under those MICs mentioned. The isolate TE1 was found to be superior on formaldehyde fixation as indicated by 93% reduction of formaldehyde in the culture medium and less accumulation of intracellular formaldehyde (0.374 mmol per gram dry weight) compared to the other two isolates. However, high level of specific activity of HPS and PHI, the key enzymes for the RuMP pathway of formaldehyde fixation was observed among the three bacterial isolates studied. Identification of those three thermotolerant methylotrophic bacteria was performed by conventional method as well as molecular biological method. Results indicated that the isolate SB4, TE1 and TB2 are closely related to the bacteria Bacillus sporothermodurans, Bacillus badius and Brevibacillus choshiensis, respectively.

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