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# Cultivable proteolytic bacteria and their proteases in three Antarctic freshwater lakes

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Antarctica is one of the extreme environments for creatures. However, in the Antarctic coastal freshwater lakes, the water temperature increases to 5-10°C in summer, and the biological activity is accelerated. In this environment, cold active protease-producing bacteria as decomposer may play a large role to hydrolyze protein produced by a kinds of protists. In this paper, we report the results of cultivation of proteolytic bacteria derived from three Antarctic coastal freshwater lakes. The water samples including surface sediments were collected from Lake Yukidori-Ike, Hotoke-Ike and Skallen-Oike during December 2012 through January 2013 in the 54th Japanese Antarctic Research Expedition. The 0.1 mL of the water samples were spread onto LB- or MBSY-skim milk agar plates and incubated at 4 °C . The colonies appeared with clear zones, indicating protease activity, were purified by single colony isolation. The purified strains were identified by the partial 16S rDNA sequences. The crude enzymes were recovered from pure cultures of 18 representative strains, and their specific activities and sensitivity to the several protease inhibitors were examined. Total 63 strains were isolated as cold-active protease producing bacteria, and grouped into seven genera, *Flavobacterium*, *Pseudomonas*, *Arthrobacter*, *Psychrobacter*, *Cryobacterium*, *Hymenobacter* and *Polaromonas*. Some of the isolates assumed to be novel species. No species was isolated from more than two lake among the three lakes. A half of the representative strains were psychrophilic and did not grow above 25 °C . From the results of inhibitor tests, nearly all of the isolates secreted metalloproteases. These results expanded our knowledge about bacterial protein degradation in Antarctic freshwater lakes.

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