

Piezophiles in Japan Sea -Relations between Japan Sea and Other ocean from the Biogeography of Piezophiles-

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Japan Sea is believed as a separated ocean from the other ocean area, like Pacific Ocean, Eastern Chinese Sea, and etc. Because of the connection sites (strait or channel) between Japan Sea and other oceans are pretty shallow, and the deepest channel is just less than 170 m, however deepest bottom of Japan Sea is more than 4,000 m. Thus people considered that the deep-sea animals would be not able to mix each other, so deep-sea bottom of Japan Sea might be perfectly separated with other oceans, even in the deep-sea biological diversity. To confirm this hypothesis, we organized the cruise, YK03-05 (principle investigator, Takako Sato) on July, 2003, to obtain the deep sediment samples from the bottom of Japan Sea, and investigated to study the diversity of deep-sea high-pressure adapted microbes, called piezophiles, in those sediment samples.

The deep sediment samples were collected from the depths of 3,000~3,100 m at Shiribeshi Trough, and keep them at liquid nitrogen conditions until use. For isolation of the piezophilic bacteria, the DEEPBATH system (deep-sea high-pressure microorganisms isolation and cultivation system, JAMSTEC) was used. The microbial community in the deep sediment samples obtained from the bottom of Japan Sea changed during the high-pressure cultivation process using the DEEP-BATH system, and based on the results of t-RFLP analysis only the genera *Moritella*, *Shewanella* and *Psychromonas* grew under such conditions (30 and 50 MPa). Continual cultivation was performed three times at 30 and 50 MPa and 4C. The population of genus *Shewanella* and *Moritella* increased during continual cultivation under both pressure conditions. The *Shewanella* sp., *Moritella* sp. and *Psychromonas* sp. were isolated from both third-cultivation mixtures, and the 16S rDNA sequences were determined from DNAs extracted from the isolated microbes. The phylogenetic relations of those isolates suggested that they were closely related with piezophilic bacteria groups isolated from Pacific Ocean bottoms. In addition, the growth profiles of those isolates, particularly *Shewanella* sp. and *Moritella* sp., were shown as piezophilic properties, and the optimal pressure conditions of them were at 30 MPa and 20 MPa, respectively. Therefore, these results suggested that pressure-adapted microbial diversity in the deep Japan Sea could be similar to that in the deep Pacific Ocean. So, it is possible to have some connection routs between Japan Sea and Pacific Ocean deep-sea bottom (Japan Trench), probably through the subsurface water lines, etc.