B319 Specific PCR Detection of an Oil-Degrading Bacterium, *Corynebacterium* sp. IC10, Using Species-Specific Primers of 16S rDNA Sequences.

Lee, Jung Hyun, Sung Young Jung* and Sang Jin Kim
Microbiology Lab., Divsion of Marine Biology, Korea Ocean Research and Development Institute

An oil-degrading bacterium, *Corynebacterium* sp. IC10, has potential in bioremediating oil-contaminated environments. To detect this organism in the environment, specific primers for PCR were designed based on 16S rDNA sequence. Four sets of the specific PCR amplification primers were tested against closely related reference strains. The two sets of primers were specific to the species of *C. variabilis*. The sensitivity of the specific PCR detection was evaluated with purified genomic DNA from strain IC10 pure culture and from sterile sands seeded with a known number of strain IC10 and *Escherichia coli*. The PCR using the specific primer set (IC417f and IC811r) was able to detect 100 ng to 1 pg of template DNA from the pure culture and 7×10^6 to 7×10^3 CFU per gram of sand in the presence of 2×10^8 *E. coli* CFU per g of sand. In the PCR detection of non-sterile sand seeded with strain IC10, 3×10^8 to 3×10^6 CFU per gram of sand was specifically detected.

B320 Bacterial community analysis of marine sediments using terminal-restriction fragment length polymorphism (T-RFLP) of 16S rRNA genes

Seong-Young Jung, Kae Kyung Kwon, Sang-Jin Kim, Hong Kum Lee, and Jung-Hyun Lee*
Microbiology Lab., Korea Ocean Research & Development Institute

The terminal-restriction fragment length polymorphism (T-RFLP) has been applied to assess the diversity of complex bacterial communities and to compare the community structure of different environments by using automated DNA sequencer. The specific fluorescence-labeled terminal restriction fragments (T-RFs) between 39 and 839 bases corresponding known added specific ribotypes were precisely measured and the PCR conditions were optimized using in vitro model community. The T-RFLP patterns of the complex bacterial communities in marine sediments were different with the extent of contamination by duration of aquaculture. The results obtained by T-RFLP were verified by cloning and sequencing 16S rDNA clones from the same marine sediments. The most abundant specific T-RFs in T-RFLP of contaminated sediments were related to a symbiont of *Rimicaris exoculata* belong to epsilon subdivision of *Proteobacteria*.