Rapid and exact molecular identification of the PSP (paralytic shellfish poisoning) producing dinoflagellate genus *Alexandrium*

Choong-jae Kim[a, *], Sook-Yang Kim[a], Kui-Young Kim[a], Young-Sil Kang[a], Hak-Gyoon Kim[a], and Chang-Hoon Kim[b]

[a] *National Fisheries Research & Development Institute, Busan 619-900, Korea*
[b] *Department of Aquaculture, Pukyong National University, Busan 608-737, Korea*

The marine dinoflagellate genus *Alexandrium* comprise PSP producing *A. acatenella, A. angustitabulatum, A. catenella, A. fundyense, A. minutum, A. ostenfeldii, A. tamiyavanichii* and *A. tamarense.* In monitoring toxic *Alexandrium,* rapid and exact species identification is one of the significant prerequisite work, however we have suffered confusion of species definition in *Alexandrium.* To surmount this problem, we chose DNA probing, which has long been used as an alternative for conventional identification methods, primarily relying on morphological approaches using microscope in microbial field. Oligonucleotide DNA probes targeting rRNA or rDNA have been commonly used in diverse studies to detect and enumerate cells concerned as a culture-indetendent powerful tool. Despite of the massive literature on the HAB species containing *Alexandrium,* application of DNA probing for species identification and detection has been limited to a few documents.

DNA probes of toxic *A. tamarense, A. catenella* and *A. tamiyavanichii,* and non-toxic *A. affine, A. fraterculus, A. insuetum* and *A. pseudogonyaulax* were designed from LSU rDNA D1-D2, and applied to whole cell-FISH. Each DNA probes reacted only the targeted *Alexandrium* cells with very high species-specificity within *Alexandrium.* The probes could detect each targeted cells obtained from the natural sea water samples without cross-reactivity. Labeling intensity varied in the growth stage, this showed that the contents of probe-targeted cellular rRNA decreased with reduced growth rate. Double probe TAMID2S1 achieved approximately two times higher fluorescent intensity than that with single probe TAMID2. This

double probe did not cross-react with any kinds of microorganisms in the natural sea waters. Therefore we can say that in whole-cell FISH procedure this double DNA probe successfully labeled targeted *A. tamiyavanichii* without cross-reaction with congeners and diverse natural bio-communities.