A Preliminary Study for Development of a Bioassay Protocol Using the Sperm of a Starfish, *Asterias amurensis*

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Bioassays using gametes of sea urchins are widely used in ecotoxicological assessments of marine environments. Since most of sea urchin species in Korean coastal water spawn from spring to autumn, bioassay with them during the winter is impossible. In the course of developing standard methods for bioassays with Korean species, we found a winter-spawning starfish, Asterias amurensis. Since reproductive mode of asteroids is similar to echinoids, the bioassay protocol for sea urchins could be applied similarly to the starfish. Here, we tested and determined several conditions for the acceptability of bioassay with A. amurensis. The least required time for formation of fertilization membrane of fertilized eggs to be easily distinguished from unfertilized ones was 60 min. The threshold of sperm to egg ratio that could make acceptable fertilization rates in controls was 3000. The allowed time for manipulation of sperm after dilution in seawater was at most 3 hr. The optimal exposure time of sperms when the response against toxicant solution was relatively stable was in the range of 20-60 min. The tolerance range of sperms to the salinity of test solution was 26-38 psu. The sensitivity of A. amurensis sperm was intermediate among marine organisms commonly used in aquatic toxicity tests. The sperm bioassay with A. amurensis can be satisfactorily applied to toxicity assessments of marine environments.

Keywords: Starfish, Asterias amurensis, Sperms, Bioassay, Optimal conditions