

## Changes in reactive oxygen species (ROS) and anti-oxidant parameter in the Manila clam, *Ruditapes philippinarum*, in relation to parasite infection and starvation

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### Introduction

Recently, mass mortality of the Manila clam has occurred since early 1990's along the west and south coasts of Korea. It is believed that parasitic infection and degradation of environmental condition are associated with the mortality of the clam species. The exposure of marine mollusks to environmental changes has been shown to induce oxidative stress through the formation of reactive oxygen and nitrogen species (ROS/RNS) and lipid peroxidation. To protect against oxidative stress, organisms possess antioxidant defenses. Therefore the antioxidant status has been suggested as a useful tool in assessing the risk of oxidative damage due to the dynamic imbalance between the antioxidant defense and pro-oxidation conditions in animals exposed to stress. In the present study, changes in oxidative stress and anti-oxidant parameter in the Manila clam, *Ruditapes philippinarum*, in relation to parasite infection and starvation were evaluated.

### Materials and Methods

Manila clams were obtained from two sites; Gomso Bay located on the west coast of Korea where the protozoan parasite *P. atlanticus* is extremely epidemic, while clams in Gimyoung on Jeju Island is *Perkinsus*-free. Samples were acclimated to tanks for a week, and then each group was divided into 2 sub-groups, feeding and non-feeding. In each tank (39 x 30 x 22 cm), 40 clams were placed and filtrated seawater was supplied daily during experiment. For feeding groups, concentrated marine algae were fed daily at the concentrations of 900 million *Isochrysis* sp. cells and 660 million *Paolova* sp. cells/30 l (Reed Mariculture Inc., USA). For non-feeding groups, filtrated seawater only was supplied daily. On

days of 0 and 15, *Perkinsus* parasitism, condition index (CI), ROS, and free radical scavenging activity were evaluated from 10 clams in each group.

## Results and discussion

Mean CI of Gimyoung clam was much lower than that of Gomso clams. No *P. atlanticus* was observed in the clams from Gimyoung, while clams from Gomso Bay contained more than 600,000 *Perkinsus* cells/gill in the beginning of the experiment. The infection intensity did not change until the end of the experiment, and no difference of infection intensity was observed in both feeding and non-feeding groups. Overall, ROS was higher in clams from Gomso Bay, irreversibly anti-oxidant was higher in the clams from Gimyoung during whole experiment. However, when starved, a negative correlation between *Perkinsus* infection and ROS generation, and a positive correlation between *Perkinsus* infection and anti-oxidant were observed in non-feeding group of Gomso Bay. These results indicate that *Perkinsus* inhibits production of ROS by the secretion of anti-oxidant as an invading mechanism in physiologically weakened host.

## 감사의 글

이 연구는 "한일 국제 공동연구"의 일환으로 한국과학재단의 연구비 지원에 의해 이루어졌습니다.