嘉南藥理學院專題研究計畫成果報告

有機氯污染物在豐年蝦之存活與成長影響
Growth and Survival Effects on Brine Shrimp (*Artemia salina*) by Organochlorine Pollutants

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ABSTRACT

Three PCB concentrations, 0 ppb, 50 ppb and 500 ppb, were prepared in the algal culture and fed to *Artemia* (brine shrimp) larvae for examing the degrees of growth effects on the larvae. Resutls show that growth conditions (length and larva/mL) of *Artemia* when fed with *Nannochloropsis* under PCB environments had stronger inhibition effects compared to that of the control group ($p < 0.05$).

Keywords: brine shrimp, *Artemia salina*, PCB, length, growth.

INTRODUCTION

Organochlorine pollutants, such as polychlorinated biphenyls (PCBs), are very stable organic chlorinated compounds and have been extensively used in industry because of their wide range of physical properties and their chemical stability. According to estimates, more than 30% of the 120 million tons of PCB produced since 1929 have been released into the open environment (Tanabe 1988). This persistent pollutant eventually finds its way to the water column and results in toxicological effects on the aquatic ecosystem of a wide variety of species ranging from phytoplankton to marine mammals (Spacie and Hamelink 1985; Phillips 1994). Their toxicity to aquatic biota is further enhanced by their ability to bioaccumulate and biomagnify within the food chain due to extremely high liposolubility (Clark and Mackay 1991; van Sprang *et al.* 1991). Serving as a valuable food sources for the higher trophic level, phytoplankton plays a very important role in supporting the growth of many aquatic biota. Once in the water column, the lipophilic PCBs partition into the more nonpolar compartments of the aquatic ecosystem or are physically absorbed in particulate matters. However, the occurrence of organochlorine residues in aquatic organisms of the food chain usually starts with the first link of the marine phytoplankton. The importance of this process is that it may cause a biomaginfication or the growth effects of the hydrophobic organic compounds to other higher trophic levels of aquatic biota.

In the present study, two common microalgae species found in local aquaculture, *Nannochloropsis oculata* and *Isochrysis galbana*, were therefore used in this short-term ecosystem and fed to an aquatic zooplankton, brine shrimp (*Artemia* sp.). Growth effects of these aquatic biota were measured under the culture with three different PCB concentrations.

MATERIALS AND METHODS

A purity >99.0% of 2,2',4,4'-tetrachlorobiphenyl (an isomer of PCBs, purchased from Ultra Scientific, USA) was weighed and dissolved in a series of dilution with acetone to achieve 100 ppb as standard stock in the present study. A salinity of 15‰ Esdschreiber medium was prepared. The prepared artificial medium was then sterilized at 121ºC for 20 min and cooled to room temperature for the culture of microalgaes. Two pure culture of microalgaes, *Nannochloropsis. oculata* and *Isochrysis galbana* were provided by the Institute of Fisheries Science, National Taiwan University. *Artemia* nauplii were obtained by hatching the cysts (*Artemia salina*, San Francisco Bay Brand) in filtered seawater (30‰ salinity) under strong aeration for 20 hr.

At the log phase of the growth curve, two PCB concentrations (50 and 500 ppb) tested in this study were prepared by pipetting 0.5 mL and 5 mL of 100 ppb stock standard to each 1 L of *Nannochloropsis* and *Isochrysis* culture. The control groups (0 ppb) were also set up by pitetting the same volume of acetone as the contaminated groups on each of the culture. Freshly hatched *Artemia* nauplii were then filtered and evenly divided into three portions to different *Nannochloropsis* culture groups with three PCB concentrations. Six hard shell clam were randomly picked up and put into each of 0 ppb, 50 ppb and 500 ppb of PCB treated *Isochrysis* culture. During the 4-d period, the length (μm) and survival numbers of nauplii (larva/mL) were measured daily. Three different PCB concentration treatments on the *Artemia* larvae were conducted for two replicates to obtain average values. Significant differences between sample means were determined with Student's *t* test.

RESULTS AND DISCUSSION

The growth effect on the length of *Artemia* nauplii during the 4-d period under different PCB conditions in *Nannochloropsis* culture is shown on Figure 1. The control group of *Artemia* had an significant increase of length on the 4-d experiment compared to the other two PCB groups ($p < 0.05$). The highest average length increases were found in the control group, from 751 μm at 1-d to 960 μm at 4-d compared to other two contaminated groups at each day. The 500 ppb PCB group caused the lowest average length increases, from 751μm at 1-d to 810 μm at 4-d. An elevated level of PCB concentration seems consistent with the growth inhibition on *Artemia* for the first 4-d period. The steady length increases for both PCB groups after 3 d suggest that the two PCB treated cultures apparently exhibited growth reduction on *Artemia* compared to the control group. For the survival conditions of the nauplii as shown in Figure 1, we found that the control group caused the higher number of *Artemia* compared with the other two PCB groups on each day ($p < 0.05$). As a lower number of zooplankton found in an enclosed aquatic environment, a higher biomass of the algae could be observed under normal conditions. The lowest number of *Artemia* nauplii together with the lowest relative biomass of *Nannochloropsis* was found under the 500 ppb PCB environment over the 4-d period. Hence, we can conclude that 500 ppb PCB could cause apparent growth reduction and toxic effects on *Artemia* by feeding with contaminated *Nannochloropsis* compared with the other two groups.

The present study reveals that the various pollutant phenomenon is a significant factor causing the growth effects on aquatic biota. Apparently, higher PCB levels play a higher growth reduction from lower phytoplankton to its predator, zooplankton.

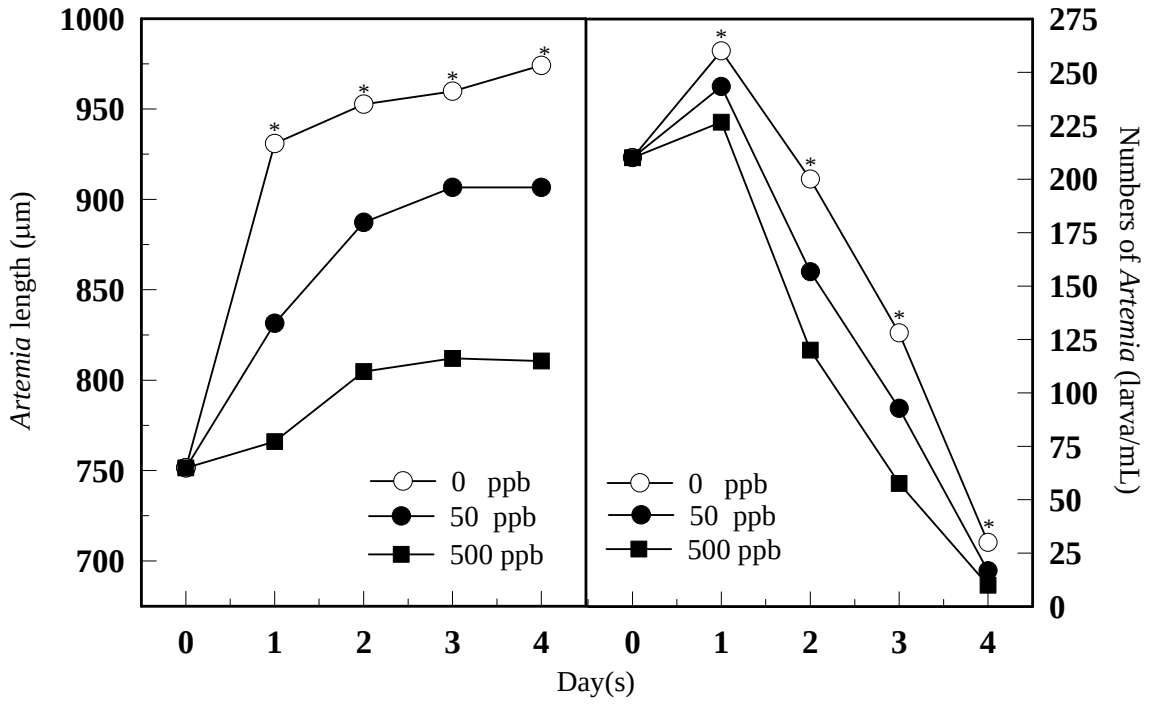

Fig. 1. Average *Artemia* length (μm) and numbers of nauplii (larva/mL) in the culture of *Nannochloropsis oculata* under three different PCB concentrations (0, 50 and 500 ppb) for 4 d. Significant differences on control group and each PCB contaminanted group in the same day are indicated by * ($p < 0.05$).

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