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Short communication

Effect of algae density on breathing and feeding of filter-feeding silver carp (*Hypophthalmichthys molitrix* Val.)

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ABSTRACT

The respiratory process of silver carp, a typical filter-feeding fish, works in conjunction with its feeding mechanism when it filters plankton in water. In the present study breathing and feeding of silver carp were measured in response to increases of algae density from 0 to 242 mg/L in order to explore the relationship between breathing and feeding in higher algae biomass environments. The results showed that (1) the oxygen consumption rate (VO_2) of the fish increased significantly with increases in algae density ($P < 0.05$), but there were no significant differences in VO_2 among fish at algae densities of 63.3–242 mg/L ($P > 0.05$). The respiratory frequency (f_R), gill ventilation (V_G) and V_G/VO_2 of the fish did not show significant differences among algae densities of 0–23.8 mg/L ($P > 0.05$). However, when algae density increased to 63.3 mg/L, the f_R, V_G and V_G/VO_2 increased significantly and reached a peak, but then declined significantly with further increases in algae density ($P < 0.05$). With increases of algae density, oxygen extraction efficiency (EO_2) first declined and then increased, with the lowest value occurring at an algae density of 63.9 mg/L. The EO_2 was negatively correlated to V_G in the present study ($P < 0.05$). There were no significant differences in respiratory stroke volume ($V_{S,R}$) among fish at algae densities of 0–242 mg/L ($P > 0.05$). (2) The filtration rate (FR) of silver carp increased significantly with increases of algae density, but did not show significant differences at levels of 63.3–242 mg/L ($P > 0.05$). The changes of the clearance rate (CR) and filtering efficiency (E) of the fish showed the same trend, in which the highest values occurred at algae densities of 63.3 mg/L and 23.8 mg/L, respectively ($P < 0.05$). In addition, an anti-filtering response occurred at algae densities over 138 mg/L. The present studies indicate that in order to acclimate to environments with higher algae biomass, silver carp can actively reduce clearance rate through a decline of filtering efficiency and/or gill ventilation.

1. Introduction

The silver carp (*Hypophthalmichthys molitrix* Val.) belongs to Cyprinidae, Cypriniformes, and occurs naturally from Heilongjiang River to Hong Song River in Asia (Li and Fang, 1993). It has received much attention worldwide because of its importance as a worldwide aquaculture species, as well as its potential for the bio-manipulation of plankton communities, especially in nutrient rich waters where blooms occur (Fukushima et al., 1999; Ke et al., 2007; Starling, 1993; Tucker, 2006; Xie and Liu, 2001; Yan et al., 2009).

Like other filter-feeding fish, the silver carp uses gills equipped with gill rakers to feed and respire (Li and Dong, 1996; Liu et al., 1992). It exhibits passive size-selection for food particles in water, but it cannot actively select for its preferred species of plankton which are evenly distributed in the water. However, the silver carp can select feeding areas where there are different plankton species or densities (Dong and Li, 1994; Dong et al., 1992). In large waters, like lakes and reservoirs, phytoplankton species are distributed in certain patterns (Reynolds, 1984), so silver carp in these waters may swim actively to specific areas to feed. Phytoplankton species are distributed evenly in fish farming ponds, especially in those with aeration facilities. However, phytoplankton communities bloom and collapse frequently in nutrient rich ponds, and as a result, cultured silver carp experience regular phytoplankton density fluctuations.

The breathing mechanism of silver carp works in conjunction with its feeding mechanism when it filters water containing plankton. Zhao et al. (2011) reported that silver carp can acclimate to lower dissolved oxygen content by regulation of respiratory and filter-feeding parameters, such as respiratory frequency, oxygen consumption rate, and gill ventilation. It is of critical importance to know how silver carp acclimate to conditions of high phytoplankton density, and regulate food intake, especially under conditions of low oxygen and high plankton density. In order to clarify the relationship between respiration and food intake under conditions of higher plankton densities, the interaction of breathing and feeding mechanisms was studied in response to different algae densities. The present results are useful for drafting stocking strategies

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of silver carp for integrated aquaculture, as well as the bio-manipulation of plankton communities in nutrient rich waters.

2. Materials and methods

2.1. Experimental materials

Silver carp (*H. molitrix* Val.) with a body weight of 61.2 ± 8.2 g and a body length of 16.6 ± 0.5 cm were obtained from a fish farm in Qingdao, Shandong Province, P. R. China. Fish were kept in an outdoor pool (15 m × 20 m) for at least 1 month prior to experiments. During this period, the fish filtered natural plankton in the pool (the main species were Chlorophyta; densities were 20–60 mg/L). The water temperature in the pool was 23 ± 2 °C; pH value was 7.5 ± 0.2.

Healthy fish from the pool were transferred into a 200 L indoor cylindrical tank for acclimation. During the acclimation period, aeration was provided continuously, and 90% of the water (dechlorinated tap water) in the tank was replaced daily. A mixture of phytoplankton at a density of 10–20 mg/L was fed daily at 09:00 and 17:00. The water in the tank was maintained at a temperature of 23.5 ± 0.6 °C. Dissolved oxygen content was maintained above 7.0 mg/L, and the photoperiod was 12L:12D. The fish were acclimated for 7 days before experiment.

In this experiment, the food organism was *Padorina morum* (diameter 20.1 ± 4.6 μm), which was obtained from the Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan, P. R. China. *P. morum* was cultured in SE medium. Phytoplankton solution of *P. morum* was filtered through the funken (mesh diameter 9.8 μm) in order to avoid toxicity of the culture medium to the experimental fish. The filtered phytoplankton particles were re-suspended in dechlorinated tap water for later use.

2.2. Experimental methods

After acclimation, silver carp were starved for 48 h, and then each fish was transferred to an individual perspex flow-through respiratory chamber (Dong et al., 1992) for parameter measurement. Each fish was acclimatized in the respiratory chamber for at least 5–6 h before measurement, until the respiratory frequency fell to resting level and stabilized. During the acclimation period, clean water without phytoplankton flowed through the respiratory chamber continuously at approximately 9 L/h (Zhao et al., 2011). The differences between the DO concentrations of the inlet and outlet water of the respiratory chamber were kept at approximately 10–15% (Hughes et al., 1983).

The inlet and outlet water of the respiratory chamber were obtained by siphoning into a glass container, following which the DO concentrations ($C_{in}O_2$ and $C_{out}O_2$, mg/L) and algae densities were measured. The DO concentrations of inspired (C_iO_2, mg/L) and expired (C_eO_2, mg/L) water, were continuously measured by siphoning water into a glass container from fixed polyethylene catheters (3 mm in inner diameter) anterior to the mouth of the fish and the outlet of opercular cavity of the fish (Zhao et al., 2011). The catheters were fixed close to the front of the mouth and the outlet of opercular cavity of the fish, respectively, but did not affix to the fish. The average flow rate through the respiratory chamber was 9.0 L/h. The respiratory chamber was covered with black cloth to avoid any visual disturbance to the fish during the measurement. Each fish in the experiment always underwent the same experimental sequence.

After blank samples (without algae in water) were measured, algae densities were increased to 5.88 mg/L, 10.6 mg/L, 23.8 mg/L, 63.9 mg/L, 138 mg/L, and 242 mg/L, respectively. Each experimental algae density was maintained for 40 min before the measurement.

There were 12 replicates in the experiment. During the experiment, the water was maintained at a temperature of 23.6 ± 0.3 °C. Because of natural sedimentation of algae cells, blank samples (without fish) were set up and measured, in order to eliminate systematic error due to sedimentation.

DO concentrations were measured with a YSI BOD probe (Model 5010) connected to an YSI oxygen meter (Model 5000-230 V, YSI Incorporated, Yellow Springs, OH, USA). Algae densities were measured under a microscope using an algae count box, and algae weights were estimated from approximate geometric volumes of *P. morum*.

2.3. Parameter measurement

Oxygen consumption rate was measured by means of a flow-through respirometry system, concomitantly with the determination of gill ventilation, respiratory stroke volume, and dissolved oxygen extraction from water according to the modification of Fernandes and Rantin (1989).

Oxygen consumption rate (VO_2, mg/kg/h) was calculated as:

$$VO_2 = V_R(C_{in}O_2 - C_{out}O_2)/W_t,$$

where V_R was the volume flow rate through the respiratory chamber (L/h), and W_t was body weight of the fish (kg).

Gill ventilation (V_G, L/kg/min) and oxygen extraction efficiency (EO_2, %) were calculated as:

$$V_G = VO_2/60(C_iO_2 - C_eO_2)$$

$$EO_2 = 100(C_iO_2 - C_eO_2)/C_iO_2.$$

Respiratory stroke volume ($V_{S.R}$, ml/kg/breath) was calculated as:

$$V_{S.R} = 1000\,V_G/f_R,$$

where f_R was respiratory frequency or filtration frequency (breaths/min), which was measured by counting the number of buccal movements of the fish during 5–8 min at each algae density level.

Filtration rate (FR, mg/kg/h), clearance rate (CR, L/kg/h), and filtering efficiency (E, %) were calculated as (Turker et al., 2003a,b; Savina and Pouvreau, 2004):

$$FR = V_R(C_{in} - C_{out})/W_t$$

$$CR = V_R(C_{in} - C_{out})/C_{in}W_t$$

$$E = 100\ \ CR/V_G$$

where C_{in} (mg/L) and C_{out} (mg/L) were the algae particle concentrations at the inlet and outlet of the respiratory chamber.

2.4. Statistical analysis

Statistical analyses were performed using SPSS 17.0 for Windows. To analyze possible differences among algae densities, a one-way ANOVA followed by Duncan's multiple comparisons test was employed after previous determination of normality and homoscedasticity of the data. Differences were considered significant at the level $P < 0.05$.

3. Results

The breathing and feeding parameters of silver carp at different algae densities are shown in Fig. 1. The respiratory frequency (f_R) and gill ventilation (V_G) of silver carp in the blank sample (without algae in water) were 42 breaths/min and 1.11 L/kg/min, respectively (Fig. 1-A; -B). With increases of algae density, f_R and V_G of the fish increased gradually, and reached a peak at an algae density of 63.3 mg/L ($P < 0.05$), and then declined significantly with further increase in algae density ($P < 0.05$). Oxygen extraction efficiency (EO_2) of the fish