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Short Communication

Physiological responses to swimming fatigue of juvenile white-leg shrimp *Litopenaeus vannamei* exposed to different current velocities, temperatures and salinities

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Swimming performance is one of the crucial factors determining the lifestyle and survival of Penaeid shrimps. This study examined under controlled laboratory conditions, the physiological responses to swimming fatigue of juvenile white-leg shrimp *Litopenaeus vannamei* (8.85 ± 0.05 cm TL) exposed to different current velocities, temperatures and salinities factors which have been correlated with their swimming performance. The swimming endurance of juveniles decreased as current velocity increased from 5.41 to 11.47 cm s^{-1} at any of the temperatures and salinities tested. Exercise to fatigue led to severe loss of serum total protein concentration (PC) and serum glucose level (SG) in *L. vannamei* exposed to different current velocities, temperatures and salinities ($P < 0.05$). Moreover, decrease of PC and SG in fatigued shrimp varied with current velocity, temperature and salinity. The results showed that the mobilization of protein and glucose in response to swimming fatigue was rapidly diminished and suggest how physiological responses to swimming fatigue of juvenile white-leg shrimp *L. vannamei* exposed to different current velocity, temperature and salinity may determine their swimming performances.

Key words: *Litopenaeus vannamei*, swimming fatigue, current velocities, temperatures, salinities.

INTRODUCTION

One of the crucial factors determining the lifestyle and survival of Penaeid shrimps is swimming performance, which may be influenced by current velocity, temperature and salinity (Dall et al., 1990; Zhang et al., 2007). While information on physiological response to swimming fatigue of Penaeids from laboratory studies may be correlated with various environmental factors, no clear cause and effect relationship has been established. The aim of this study was to examine under controlled laboratory conditions, the physiological responses to swimming fatigue of juvenile white-leg shrimp *Litopenaeus vannamei* exposed to different current velocities, temperatures and salinities factors which have been implicated in swimming performance of Penaeid shrimps.

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MATERIALS AND METHODS

Experimental animals

The white-leg shrimps, *L. vannamei* used in the experiment were obtained from local shrimp farms (Shazikou, Qingdao). Only shrimps in the intermolt stage were used for the study. The shrimps were acclimated in a 2 m^3 recirculating fiberglass tank for two weeks before experiments commenced. During this acclimation period, shrimps were fed twice daily with commercial pellets (HaiMa Ltd,

FuJian, China) at 5% of their body mass. The water in the tank was maintained at 20 ± 0.5°C, salinity of 32.0 ± 1.0‰, dissolved oxygen (DO) of 6.30 ± 0.50 mgl^{-1}, photoperiod of 14 L / 10 D, and pH of 8.20 ± 0.118. Water was exchanged at the daily rate of 5% tank volume with 1 µm filtered and ultraviolet sterilized seawater.

Experimental apparatus

Trials were conducted in a large annular flume of 150 L total volume, which comprised of a rectangular swimming channel of 40 × 15 × 14 cm (length × width × height), and a motor-driven propeller (LifeTech Model AP5600) which delivers water at a rate of 90 L min^{-1} (for schematic diagram of the rectangular swimming channel see Zhang et al., 2006).

The mean current velocities of 5.41 ± 0.12, 6.78 ± 0.15, 8.21 ± 0.11, 10.11 ± 0.17 and 11.47 ± 0.20 cms^{-1} (mean±S.D.) were chosen based on several pilot trials, in which all test shrimps failed to swim for more than 9000 s. The duration of 9000 s was prescribed by referring to the upper end time for endurance test described by Zhang et al. (2006).

Experimental design

The swimming performance trials were conducted at water temperatures of 15, 20 and 25°C, and salinities of 15, 32 and 40‰. Water temperature was maintained by the combined use of a cooling and a heating system. The desired salinities were obtained by diluting seawater with fresh water, or by adding artificial salt.

In each water temperature or salinity treatment, 100 individuals (8.85±0.05 cm TL and 4.13 ± 0.017 g wet body mass) were randomly size selected from those held in rearing tank and were then acclimatized to the desired water temperature or salinity level by a procedure described by Chen et al. (1995). After acclimation, shrimp were divided into five groups of 20 individuals. One group was tested at each water flow velocity.

Shrimps were fasted for 12 h before been used for swimming trials. Two shrimp were used together in each swimming trial. The shrimps were allowed to acclimatize to the swimming channel for 4 h before testing. They were trained for 15 min at a low speed of approximately 4 cms^{-1} to orient them to the current and then accelerated over 2 min period to the desired current velocity. When a shrimp showed fatigue, it was immediately removed from the flume and surface-dried on a piece of absorbent paper. Then samples of hemolymph were with- drawn immediately according to the procedure described later. The criterion for swimming fatigue was established and a shrimp was considered fatigued when it fell against the downstream screen and could not resume swimming after three manual prods at the tail. No feed was given during the experimental period.

Seawater used in the experiment was prepared a day before the commencement of experiment by composite sand filtration and ultraviolet sterilization. The tank was continuously aerated and the DO was maintained at >6.40 mgl^{-1}. The tank was illuminated by two 60 W lights, one at each end, to ensure uniform illumination.

Hemolymph collection and biochemical assessment

To investigate the physiological response to swimming fatigue, serum total protein concentration (PC) and serum glucose level (SG) were measured before swimming and immediately following exhaustive exercise. In order to approximate initial PC and SG in fatigued shrimp, pre-exercise hemolymph samples were taken from 20 individuals of the same size as test shrimp before each test (control).

Hemolymph sample was collected individually by inserting a 26-gauge needle attached to a 1 ml syringe into the pericardial sinus through the intersegmental membrane which is between the cephalothorax and the abdominal segment, and transferred to a 2 ml microcentrifuge tube, and allowed to coagulate for 2 h at room temperature and overnight at 4°C. The clot was broken up using a pipette tip and then repeatedly centrifuged at 3000 × g in a microcentrifuge for 15 min to obtain the serum, which was analyzed immediately. PC of the shrimp was determined using the Biuret Protein Assay kit (Nanjing Jiancheng Bioengineering Institute, Nanjing, China) with bovine serum albumin as protein standard and was expressed as mg ml^{-1} of animal serum. The SG was measured with a commercial kit (GOD-PAD, Zhongsheng Beikong Bio-Technology and Science Inc., Beijing, China) using 10 µl of serum samples and was expressed as mMol L^{-1} of animal serum.

Data analysis

Experimental results were analyzed by two-way analysis of variance (ANOVA). When ANOVA indicated a significant difference between factors, Duncan's multiple range tests were applied. PC and SG of pre- and post-exercise shrimp were analyzed using independent sample t-tests. These statistical analyses were performed using SPSS 11.5 statistical software and the significance level was $P < 0.05$ for all analysis.

RESULTS AND DISCUSSION

Serum total protein can serve as a significant source of metabolic energy for crustaceans (Claybrook, 1983; Mugnier and Justou, 2004) and therefore has been used to provide a rapid and non-destructive means of assessing an animal's physiological condition (Moore et al., 2000). The mobilization of glucose in response to stress is generally accepted as a means of providing extra energy resources, enabling the animal to overcome the disturbance (Pascual et al., 2003; Acerete et al., 2004).

Data in this study indicated that the swimming endurance decreased as current velocity increased from 5.41 to 11.47 cms^{-1} at any of the temperatures and salinities tested. Exercise to fatigue led to severe loss of PC and SG in *L. vannamei* exposed to different current velocities, temperatures and salinities ($P < 0.05$) (Tables 1 and 2), showing that the mobilization of protein and glucose in response to swimming fatigue was rapidly diminished or that all the circulating blood metabolites were rapidly used (Pascual et al., 2003).

Moreover, results in this study showed that the decrease in PC and SG in fatigued shrimp varied with current velocity, temperature and salinity. Therefore, current velocity, temperature and salinity may limit the swimming performance of *L. vannamei* by affecting the PC and SG levels.

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Table 1. Physiological responses of *L. vannamei* after swimming fatigue at different temperatures and current velocities (means±S.E, n = 20).

Physiological parameter	Temp. (℃)	Current velocity (cms^{-1})					
		Control	5.41	6.78	8.21	10.11	11.47
Serum total protein (mg ml^{-1})	15	67.3±0.6^{a}	63.7±0.3^{a*}	57.5±0.4^{a*}	55.9±0.4^{a*}	56.3±0.3^{a*}	54.1±0.4^{a*}
	20	79.2±0.9^{b}	73.9±0.3^{b*}	73.8±0.2^{b*}	59.9±0.6^{b*}	42.5±0.3^{b*}	35.4±0.3^{b*}
	25	76.6±0.6^{c}	62.9±0.2^{c*}	55.5±0.2^{c*}	62.1±0.2^{c*}	58.2±0.3^{c*}	59.8±0.2^{c*}
Serum glucose level (mMol L^{-1})	15	0.97±0.04^{a}	0.94±0.001^{a}	0.81±0.003^{a*}	0.82±0.004^{a*}	0.83±0.002^{a*}	0.73±0.009^{a*}
	20	1.09±0.90^{b}	0.86±0.013^{b*}	0.91±0.013^{b*}	0.96±0.008^{b*}	0.88±0.013^{b*}	0.66±0.011^{b*}
	25	1.09±0.02^{b}	0.85±0.011^{b*}	0.85±0.007^{c*}	0.82±0.006^{a*}	0.88±0.012^{b*}	0.81±0.015^{c*}

* Indicates a very significant difference from control (P < 0.01); different letters in the same column are significantly different (P < 0.05).

Table 2. Physiological responses of *L. vannamei* after swimming fatigue at different salinities and current velocities (means±S.E, n = 20).

Physiological parameter	Salinities (‰)	Current velocity (cm s^{-1})					
		Control	5.41	6.78	8.21	10.11	11.47
serum total protein (mg ml^{-1})	15	65.9±1.3^{a}	56.4±3.6^{a*}	52.6±3.9^{a**}	61.9±1.5^{a}	44.2±1.9**	48.9±2.6**
	32	79.2±0.9^{b}	71.5±1.1^{b**}	69.9±1.5^{b**}	58.8±1.1^{ab**}	44.9±2.2**	42.7±3.1**
	40	66.1±0.9^{a}	45.6±1.4^{c**}	49.9±0.7^{a**}	54.4±2.1^{b**}	49.9±2.3**	48.1±2.7**
Serum glucose level (mMol L^{-1})	15	1.07±0.03	0.77±0.08**	0.90±0.07^{a*}	0.90±0.06^{a*}	0.84±0.06**	0.89±0.06^{a*}
	32	1.11±0.47	0.82±0.07*	0.79±0.04^{a**}	0.71±0.06^{b**}	0.84±0.02**	1.04±0.06^{b}
	40	1.14±0.03	0.88±0.06**	1.06±0.04^{b}	0.93±0.05^{a**}	0.84±0.04**	1.16±0.02^{b}

* Indicates a significant difference from control (P < 0.05); ** indicates a very significant difference from control (P < 0.01); different letters in the same column are significantly different (P < 0.05).

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REFERENCES

Acerete L, Balasch JC, Espinosa E, Josa A, Tort L (2004). Physiological responses in Eurasian perch (*Perca fluviatilis*, L.) subjected to stress by transport and handling. Aquaculture, 237: 167-178.

Chen JC, Lin MN, Ting YY, Lin JN (1995). Survival, haemolymph osmolality and tissue water of *Penaeus chinensis* juvenile acclimated to different salinity and temperature levels. Comp. Biochem. Physiol. 110A: 253-258.

Claybrook DL (1983). Nitrogen metabolism. In: Mantel LH (Eds). The Biology of Crustacea. vol. 5. Internal Anatomy and Physiological Regulation, Academic Press, New York, pp. 162-213.

Dall W, Hill BJ, Rothlisberg PC, Staples DJ (1990). The Biology of the Penaeidae. Advances in Marine Biology, vol. 27. Academic Press, New York, pp. 17-340.

Moore LE, Smith DM, Loneragan NR (2000). Blood refractive index and whole-body lipid content as indicators of nutritional condition for penaeid prawns (Decapoda: *Penaeidae*). J. Exp. Mar. Biol. Ecol. 244: 131-143.

Mugnier C, Justou C (2004). Combined effect of external ammonia and molt stage on the blue shrimp *Litopenaeus stylirostris* physiological response. J. Exp. Mar. Biol. Ecol. 309: 35-46.

Pascual C, Sánchez A, Sánchez A, Vargas-Albores F, LeMoullac G, Rosas C (2003). Haemolymph metabolic variables and immune response in *Litopenaeus setiferus* adult males: the effect of an extreme temperature. Aquaculture, 218: 637-650.

Zhang PD, Zhang XM, Li J, Huang GQ (2006). Swimming ability and physiological response to swimming fatigue in whiteleg shrimp, *Litopenaeus vannamei*. Comp. Biochem. Physiol. 145A: 26-32.

Zhang PD, Zhang XM, Li J, Huang GQ (2007). The effects of temperature and salinity on the swimming ability of whiteleg shrimp, *Litopenaeus vannamei*. Comp. Biochem. Physiol. 147A: 64-69.