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Effects of dopamine on the immunity of white shrimp *Litopenaeus vannamei*

Winton Cheng [a], Hung-Tien Chieu [a], Chiung-Hui Tsai [a], Jiann-Chu Chen [b,*]

[a] *Department of Aquaculture, National Pingtung University of Science and Technology, Pingtung, Taiwan, 912, ROC*
[b] *Department of Aquaculture, College of Life and Resource Sciences, National Taiwan Ocean University, Keelung, Taiwan, 202, ROC*

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Abstract

The total haemocyte count (THC), phenoloxidase activity, respiratory burst, superoxide dismutase (SOD) activity, phagocytic activity and clearance efficiency in response to pathogen *Vibrio alginolyticus* were measured when the white shrimp *Litopenaeus vannamei* (20.0 ± 1.5 g) were injected individually with dopamine at 10^{-8}, 10^{-7} and 10^{-6} mol shrimp^{-1}, respectively. For the shrimp that received dopamine at 10^{-7} and 10^{-6} mol shrimp^{-1}, the THC decreased by 25% and 39%, phenoloxidase activity decreased by 15% and 32%, respiratory burst decreased by 21% and 36%, and SOD activity decreased by 50% and 63%, respectively, after 4 h. The phagocytic activity and clearance efficiency of shrimp that received dopamine at either dose decreased significantly after 2 h. The THC, phenoloxidase activity, respiratory burst, SOD activity, phagocytic activity and clearance efficiency returned to normal values after 16, 8, 8, 24, 16 and 4 h, respectively, for the shrimp that received dopamine at either dose. In another experiment, *L. vannamei* which had received dopamine at 10^{-8}, 10^{-7} and 10^{-6} mol shrimp^{-1} were challenged after 1 h by injection with *V. alginolyticus* at 1.0×10^5 colony-forming units (cfu) shrimp^{-1} and then placed in seawater of 20‰. The cumulative mortality of shrimp that received dopamine at either dose was significantly higher than that of shrimp that received saline after 8 h, and of shrimp that received saline at the termination of the experiment (48 h after the challenge). It is therefore concluded that dopamine administration at 10^{-6} mol shrimp^{-1} or less causes immune modulation of *L. vannamei*.

Keywords: *Litopenaeus vannamei*; Dopamine; Total haemocyte count; Phenoloxidase activity; Respiratory burst; Superoxide dismutase activity; Phagocytic activity; Clearance efficiency

* Corresponding author. Tel.: +886 2 2462 2192x5205; fax: +886 2 2462 0295.
E-mail address: jcchen@mail.ntou.edu.tw (J.-C. Chen).

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1. Introduction

White shrimp *Litopenaeus vannamei*, which is native to the Pacific coast of central and south America, was introduced to the Eastern hemisphere in 1985, and has since become the primary species cultured in Thailand, Taiwan, and China [1]. Since 2001, shrimp farmers have experienced disease problems causing production declines in farmed *L. vannamei*. The symptoms differ from those of TSV (Taura Syndrome Virus) in Taiwan [2]. A bacterium *Vibrio alginolyticus* isolated from the diseased *L. vannamei* with whitish musculature and inactivity is considered to be a secondary and opportunistic pathogen, and cause mortality of shrimp under ammonia stress [3,4].

In teleosts, the primary response to physiological stress involves the release of corticosteroids and catecholamines. These then induce hyperglycaemia as a secondary response [5]. Several biogenic amines which function mainly as neuroregulators (neurotransmitter and neuromodulators), including serotonin, dopamine, octopamine, histamine, noradrenaline (norepinephrine), adrenaline (epinephrine), tryptamine and tyramine have been identified and quantitatively measured in crayfish *Pacifastacus leniusculus* and other decapod crustaceans [6–8]. Among the biogenic amines, the presence of dopamine and serotonin in the crustacean nervous system is well established [9].

It is known that when Pacific oyster *Crassostrea gigas* are subjected to mechanical disturbance, noradrenaline and dopamine are released into the circulatory system, subsequently decreasing immune functions [10], and increasing susceptibility to *Vibrio splendidus* infection [11]. It is also known that when the abalone *Haliotis tuberculata* are subjected to a 15 min mechanical disturbance, noradrenaline and dopamine are released, and immune parameters such as haemocyte counts decrease, as do migratory activity, phagocytic and respiratory burst capacity of the haemocytes [12]. However, there is no knowledge on the release of biogenic amines and their potential role in immunosuppression of penaeid shrimps under stress.

Environmental stressors like hypoxia, ammonia, nitrite and Cu^{2+} have been reported to cause reduction in immune ability of blue shrimp *Litopenaeus stylirostris* [13], as well as white shrimp *L. vannamei* [3,14,15]. Dopamine has been found to mimic the action of CHH (crustacean hyperglycaemic hormone) in increasing the level of glucose in tiger shrimp *Penaeus monodon* [8]. It is assumed that penaeid shrimp under such stress may increase their levels of biogenic amines including dopamine, which subsequently leads to immunosuppressive effects and increases susceptibility to pathogen infection. Accordingly, the purpose of the present study is to examine: (1) the effect of dopamine on the susceptibility of *L. vannamei* to *V. alginolyticus*, and (2) the immune response of *L. vannamei* injected with dopamine. For the latter purpose, we examined THC (total haemocyte count), phenoloxidase activity, respiratory burst, SOD (superoxide dismutase) activity, phagocytic activity and clearance efficiency of shrimp to *V. alginolyticus*.

2. Materials and methods

2.1. *L. vannamei*

L. vannamei juveniles (18–22 g) were obtained from a commercial farm in Pingtung, Taiwan, and acclimated in the laboratory for 2 weeks before experimentation. Only shrimps in the intermoult stage were used for the study. The molt stage was determined by the examination of uropods in which partial retraction of the epidermis could be distinguished [16]. For the susceptibility experiment, test and control groups were comprised of 10 shrimps each in triplicate. For the determination of immune parameters, tests were carried out in eight replicate test groups consisting of one shrimp each in 20 l PVC tanks containing 10 l aerated test solution. In all tests, the shrimps were fed twice daily with a formulated shrimp diet (Shinta