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A Preliminary Study on the Effect of Hypoxia on Carotenoid Metabolism in Black Tiger Shrimp *Penaeus monodon* Fabricius

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The pigmentation of black tiger shrimp *Penaeus monodon* is due to astaxanthin, astaxanthin monoester, and astaxanthin diester. These carotenoids are biosynthesized from beta-carotene or zeaxanthin. Biosynthesis is postulated to be mediated by the enzymes C3 and C4 monooxygenase and caroten-4-ol dehydrogenase. Blue-shrimp syndrome, characterized by low total astaxanthin levels in shrimp epidermis, is associated with nutritional deficiency for carotenoids but other factors such as high organic matter, hypoxia, high density, and high pH may also be involved. In this study, the effect of hypoxia on carotenoid metabolism in *P. monodon* was investigated with respect to retention and transformation of carotenoids and the activity of the enzymes C3 and C4 monooxygenase and caroten-4-ol dehydrogenase.

Two tanks were prepared each containing 25 pieces of shrimp fed a control diet (not containing any carotenoid) for two weeks. Shrimps in one tank (Treatment 1) were given beta-carotene supplemented diet (3.8 ppm) in the next two weeks while those in the other (treatment 2) were fed the control diet. Two days after introduction of the test diets, both treatments were subjected to hypoxia challenge (dissolved oxygen, 2.5-2.7 mg/L) and the carotenoid content and profile of feces monitored for succeeding days. Assay of enzyme activity was performed five days after hypoxia challenge. Results showed a ten-fold increase in fecal total carotenoid content of shrimps in treatment 2 (122.2 ppm) compared with that in treatment 1 (12.4 ppm) on hypoxia challenge and restores to basal level (3.9-4.1 ppm) after three days. The carotenoids found in the feces of shrimps in treatment 2 were astaxanthin, astaxanthin monoester, astaxanthin diesters, and an unidentified carotenoid which was also found in the muscle of stressed shrimp. Using beta-carotene as substrate, conversion to astaxanthin was not observed in the hepatopancreas homogenates of shrimps in both treatments. Instead, a slow conversion of astaxanthin to beta-carotene (i.e., a reverse reaction) was noted in treatment 2 exhibiting twice the activity in treatment 1. Radiolabelling studies using $^{14}$C-labelled astaxanthin as substrate confirmed the existence of this enzyme-mediated reductive pathway from astaxanthin which occurs at a very slow rate. This study demonstrated the carotenoid-depleting effect of hypoxia on shrimp, both through enhanced fecal release and moderately elevated reductive pathway from astaxanthin. This effect suggests a possible mechanism by which blue-shrimp syndrome may develop.