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Growth performance and nutritional composition of *Hemifusus ternatanus* under artificial culturing conditions

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ABSTRACT

Juvenile *Hemifusus ternatanus* were cultured in a double-layer culturing system for 186 weeks in order to assess growth performance. The growth rate of *H. ternatanus* under artificial culturing conditions was high during the first 137 week post-hatching, followed by a period of relatively slower growth. The shell of the cultured sub-adult whelks was shorter and thicker than that of the wild individuals with same weight. The proximate, amino acid and fatty acid compositions of *H. ternatanus* foot muscles were analyzed to compare the quality of cultured and wild whelks. Both the cultured and wild *H. ternatanus* contained high levels of flavor-enhancing amino acids. No significant difference was observed in protein content, moisture content or amino acid composition. The cultured whelks contained significantly higher levels of fat and ash ($P < 0.05$). The total saturated, monounsaturated, and polyunsaturated (PUFA) fatty acids were statistically similar between the cultured and wild groups; however, there were significant differences in the content of individual of fatty acids between the two groups, including 17:0, 16:1, 18:1n-9c, 18:2n-6t, 20:4n-6, 22:5n-3, 22:6n-3, and n-6 PUFAs ($P < 0.05$). The fatty acids 21:0, 15:1, 22:1n-9, 18:2n-6c, and 18:3n-3 were detected in the cultured *H. ternatanus* only. On the contrary, 18:1n-9t was detected only in the wild *H. ternatanus*. This study provides novel information regarding the growth and composition of cultured *H. ternatanus* which will be helpful for the development of aquaculture practices for this species.

Statement of Relevance: This manuscript provides new findings which should be of interest to readers involved in the reproduction and aquaculture of gastropods. *H. ternatanus* is a marine whelk with agreeable taste and is a potential candidate species for aquaculture. Aquaculture production of *H. ternatanus* is increasing due to its high market price and the depletion of natural stocks. There is currently no information regarding the nutritional composition of cultured *H. ternatanus* compared to its wild counterpart. The results of this study will help consumers better understand the nutritional value of cultured whelks and provide aquaculturists with important information for the development of formulated feeds.

1. Introduction

Hemifusus ternatanus is a predatory marine whelk with a shell that can reach up to 40 cm in length that inhabits the area from the eastern and southern coast of China to Japan (Phillips and Depledge, 1986). These whelks are sold as a luxury food items in markets at a price that ranges from 60 to 90 euros per kilogram. Due to high market demand and prices, an increasing number of whelks are being captured by trawlers. Overfishing has resulted in the severe depletion of natural *H. ternatanus* stocks, increasing the importance of artificial breeding and aquaculture of this species (Xu et al., 2006).

An understanding of the growth performance of a species is important for the development of aquaculture practices. Although some studies have investigated prey preference (Morton, 1986a), the reproductive system (Cao et al., 2010), artificial breeding (Hong, 2010) and juvenile growth (Xu et al., 2009) of *H. ternatanus*, the culturing of this species has been limited to the larval and juvenile stages. In a previous study, *H. ternatanus* juveniles fed live clam grew at a high rate of over 1 cm per month (Tang et al., 2012a,b), but little is known about the growth of sub-adult and adult individuals.

H. ternatanus is a highly specialized predator of bivalves, similar to the related species *Hemifusus tuba*, which may be explained by its relatively stable tropical environment and predictable food supply (Morton, 1985). Under laboratory conditions, juvenile *H. ternatanus* were found to prefer live *Meretrix meretrix* (Tang et al., 2012a,b), which is expected to be different from its natural prey items. Diet has been shown to have an impact on nutritional composition and taste (Prato et al., 2010; Woodcock and Benkendorff, 2008). Besides genetic basis, the quality of seafood products is also dependent on the characteristics of environment (Orban et al., 2007). However, information regarding the nutritional composition of *H. ternatanus* cultured under artificial conditions

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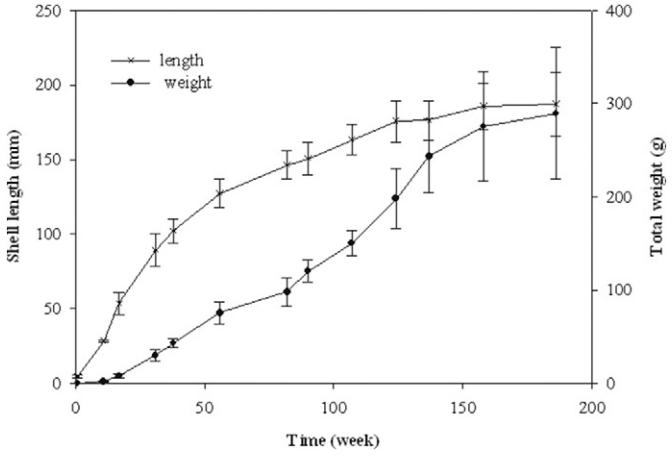

Fig. 1. The shell length and total weight of juvenile *H. ternatanus* during 186 week culturing.

is not currently available. In order to develop methods for the profitable commercial production of *H. ternatanus*, it is necessary to understand the growth and nutritional value of artificially-cultured whelks.

The aim of this research was to study the growth performance of cultured juvenile *H. ternatanus*, to assess its suitability as an aquaculture species. This research also represents the first attempt to evaluate the differences in amino acid and fatty acid profiles between wild and cultured *H. ternatanus*, which will be helpful for the development of formulated feeds for this species.

2. Materials and methods

2.1. Animals

Wild whelks were captured from the coast of Hainan Province, China, and the individuals with a shell length over 25 cm were selected for laboratory spawning. The selected whelks with a male–female ratio of 1:1 were placed in a 4.0 m × 1.5 m × 1.2 m glass fiber-reinforced plastic aquarium with 6000 L seawater at 21–23 °C and salinity at 31 psu for maintenance. After copulation, the female whelks laid egg capsules affixed to the wall of the aquarium. Juveniles were hatched and cultured as previously reported (Tang et al., 2012a,b). The egg capsules were collected and put into floating baskets for hatching. When a cleft was observed at the apex of the egg capsule (about 60 days after laying), the juveniles were released by cutting the capsules. The juveniles were cultured in floating baskets until mean shell length was 1.5 cm and were then transferred into a 6000-L aquarium.

In order to maintain water quality and reduce disturbance to whelks caused by operations such as transferring and cleaning, a double-layer flow-through culturing system was set up on the bottom of the aquarium (accounting for four-fifths of the bottom area). There is a sand layer about 15 cm above the aquarium bottom. Juveniles were cultured in the upper sand layer in the system with the density of 20 juveniles per square meter. Seawater flows into the aquarium at a constant rate of 2–3 L per minute. Air is supplied below the sand layer through PVC tubes. Up-ward aeration through the sand can remove dirty residues to prevent from the release of waste gases and keep the sand layer clean.

2.2. Diet and feeding

The juvenile whelks were fed live clam (*M. meretrix*) during the culturing in the floating baskets and the double-layer system. The clams were cut through the adductor muscles using a knife and then fed to *H. ternatanus*. Excess food was offered during one feeding time per day. Residual diet and empty clam shells were removed the next day. The juveniles were cultured for 186 weeks during which the seawater temperature ranged from 19 °C in winter to 33 °C in summer and the salinity varied between 23 and 33 psu. During the experiment, the shell length and total wet weight of the juveniles were frequently measured. Every time a total of 30 juveniles were randomly selected for measurement.

2.3. Biochemical analysis

At the end of feeding experiment, the foot muscle of cultured and newly captured wild whelks of similar total weight were sampled for biochemical analysis. All biochemical analyses were performed in triplicate and the results were presented as mean and standard deviation. The proximate and amino acid composition of the clam *M. meretrix* had been analyzed previously (Tang et al., 2012a,b).

The proximate composition of the foot muscle samples, including moisture, protein, lipid, and ash, were analyzed by standard methods (AOAC, 2005). Moisture level was measured after drying the sample at 105 °C to a constant weight. Protein content was determined following the Kjeldahl method (Foss Tecator Kjeltec 2200 analyzer, Warrington, UK). Fat content was determined by petroleum ether extraction (Foss 2043 Extraction Apparatus, Warrington, UK). Ash was determined by incineration at 550 °C in a muffle furnace.

Amino acids were analyzed following the methods outlined by Wu et al. (2010). Freeze-dried muscle samples were hydrolyzed with 6 mol L^{-1} HCl for 24 h at 110 °C. The hydrolyzate was dried under vacuum and dissolved in sodium citrate solution (pH 2.2), and filtered through a 0.45-μm Millipore nylon membrane filter. Amino acid analysis was performed using 1 μL of sample with a Biochrom 20 Amino Acids Analyzer (Biochrom Ltd., Cambridge, UK). The amino acid composition of the sample was calculated by comparison with the retention time and peak areas of the standards (Sigma-Aldrich, St. Louis, MO, USA). Amino acid concentration was presented as g/100 g dry weight.

For fatty acid composition analysis, total lipids were extracted using a chloroform-methanol solution (2:1, v/v). Fatty acid methyl esters (FAMES) were prepared from total lipids using 15% (w/v) BF3-methanol reagent according to the ISO 5509 method (ISO, 2000), and were analyzed using an HP-6890 GC gas chromatograph (Agilent Technologies Inc., USA). The fatty acid composition of the samples was determined by comparing the retention times to those of a standard FAME mixture (Supelco 37 Component Fame Mix, 10 mg mL^{-1} in methylene chloride, CRM 47885-U. Supelco, Bellefonte, PA 16823-0048, USA). Individual fatty acid content was presented as the percentage of a particular fatty acid relative to the total fatty acid content.

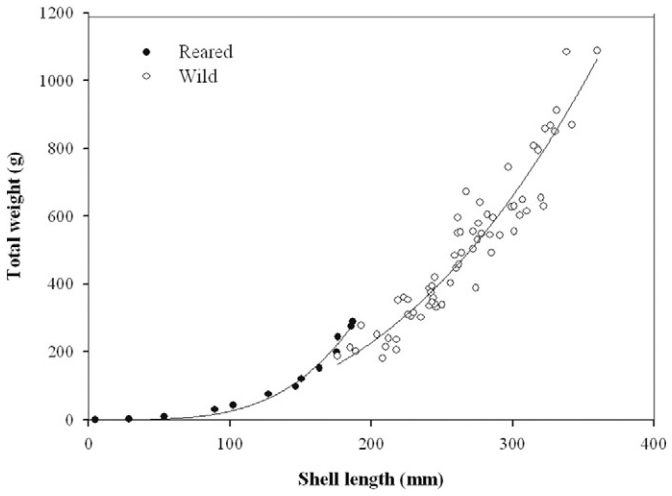

Fig. 2. Relationship between the shell length and total weight of the cultured and wild *H. ternatanus*.