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Deleterious effects of food restrictions in yellowtail kingfish *Seriola lalandi* during early development

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Abstract

The effects of delayed first feeding and food deprivation on the structure and function of the digestive system in yellowtail kingfish *Seriola lalandi* larvae and juveniles were studied through histological examinations and enzymatic analyses. The experimental design included a conventional feeding regime with initial feeding from 3 days after hatching (DAH) as a control, delayed first feeding until 5 DAH, a first 3-d food deprivation from 12 to 15 DAH, and a second 3-d food deprivation from 33 to 36 DAH. Fish samples for histological and enzymatic analyses were taken on 5, 15, and 36 DAH, respectively. The delay of first feeding and a 3-d food deprivation on 15 DAH significantly reduced the height of enterocyte cells in the midgut, but a 3-d food deprivation on 36 DAH did not significantly reduce the cell height. Lipid vacuoles and supranuclear vacuoles disappeared from the epithelial cells after the fish had experienced the delay of first feeding or a 3-d food deprivation on 15 DAH. Total and specific activities of trypsin and amylase were reduced by the delay of first feeding. The 3-d food deprivation on 15 DAH reduced specific activities of trypsin, amylase and alkaline phosphatase, and total activity of amylase, but the 3-d food deprivation on 36 DAH only reduced the amylase activity. This study indicates that yellowtail kingfish larvae are more vulnerable to starvation in the first 2 weeks after the start of first feeding but fish become more tolerant to a short-term starvation after 33 DAH. Therefore, any delay of feeding during the first 2 weeks may impair histological structure and cause malfunction of the digestive system in yellowtail kingfish larvae. Our data also suggest that the enterocyte morphology, the number of supranuclear vacuoles in the intestine and the activity of digestive enzymes can be used as indicators of the nutritional condition of fish larvae.
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Keywords: Digestive enzyme; Digestive system; Histology; Larvae; Starvation; Yellowtail kingfish

1. Introduction

Starvation is probably one of the major causes of larval fish mortality in nature (May, 1974; Margulies, 1993) and in aquaculture (Dou et al., 2002). In larval fish, even a short period of starvation after the onset of exogenous feeding can cause severe nutritional problems, leading to drastic mortality and deformity in the early stage (Kjorsvik et al., 1991). Although a plenty of live food is supplied into the culture tank, fish may still be suffering from starvation due to poor vision and mouth gape limitation (Planas and Cunha, 1999). The digestive tract of larval fish and its associated glands,

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such as liver and pancreas, are usually the most sensitive tissues affected by starvation (Theilacker, 1978; Bisbal and Bengtson, 1995). Degeneration of the digestive system caused by starvation reduces the surface area for nutrient absorption of the digestive tract and the digestive capability of re-feeding larvae (Hall and Bellwood, 1995; Gisbert et al., 2004). Furthermore, the changes in digestive tract, liver and pancreas may affect the synthesis of the digestive enzymes and functions of the digestive system (Dabrowski, 1982; Cousin et al., 1987). The ability of larval fish to withstand a short period of starvation is critical to the survival and growth of fish larvae.

Starved larvae usually exhibit a widespread histological degeneration, especially in the digestive tract and associated glands (Strussmann and Takashima, 1989; Yufera et al., 1993), and re-fed larvae cannot digest food and die even with the food in the gut (Kjorsvik et al., 1991). Consequently, starvation usually results in shrinkage of enterocytes and reduction in the height of enterocyte cells both in midgut and hindgut (Theilacker and Watanabe, 1989; Bisbal and Bengtson, 1995), lack of supranuclear vacuoles in the hindgut (Oozeki et al., 1989; Crespo et al., 2001), degeneration of the cellular structure in both liver and pancreas (O'Connell, 1976; Kjorsvik et al., 1991) and disorder of trunk musculature (Bisbal and Bengtson, 1995; Gisbert et al., 2004). In starved larvae, trypsin (Segner et al., 1989; Pedersen et al., 1990; Moyano et al., 1996), amylase (Cousin et al., 1987; Moyano et al., 1996; Kim et al., 2001), lipase (Kim et al., 2001), pepsin (Dabrowski, 1982), aminopeptidase (Cousin et al., 1987; Segner et al., 1989) and alkaline phosphatase (Cousin et al., 1987; Yufera et al., 1993) reduce to a low level compared with those in the fed larvae.

As an important commercial species, yellowtail kingfish has been cultured in South Australia for over a decade (Fowler et al., 2003). Although the larviculture techniques have been achieved by several local hatcheries for this species, high larval deformities and mortalities have hindered the industry development. Because nutritional deficiency is considered a major cause of larval fish mortality and deformity (Dhert et al., 1998), and the ability to tolerate starvation varies among fish larvae (Dou et al., 2002), it is necessary to study the changes in the histological structure and enzyme composition of the digestive system during starvation in yellowtail kingfish.

Recently, based on the early ontogeny of the digestive tract and accessory glands, Chen et al. (2006a) divided the development of the digestive system of yellowtail kingfish into three phases. The first phase is from hatching to exogenous feeding on 3 DAH, when the digestive tract is a simple tube. The second phase is from the onset of exogenous feeding to the appearance of gastric glands in the stomach, and the third developmental phase starts from the appearance of gastric glands on 15 DAH. In this study, the initial feeding was postponed to 5 DAH to examine how fish cope with the delayed feeding. The first period of food deprivation was from 12 to 15 DAH when fish larvae switch diet from rotifers to *Artemia* nauplii, and the second period of food deprivation was between 33 and 36 DAH when fish have metamorphosed and been weaned to a compound diet. The objective of this study was to evaluate the effect of delayed first feeding and a 3-d period of food deprivation at two developmental stages on the histological structure of the digestive system and major digestive enzymes in yellowtail kingfish. Determining any change of the fish digestive system due to feeding disruptions during these critical stages of fish development could provide a better understanding of the ability of these fish to tolerate starvation. The result could be useful to design appropriate feeding regimes for hatchery managers toward improvement of the culture of fish larvae.

2. Materials and methods

2.1. Larval fish rearing and sampling

Fertilized eggs of yellowtail kingfish were obtained from the Clean Seas Aquaculture Hatchery (Arno Bay, South Australia), and transported to the South Australian Aquatic Sciences Centre, Adelaide. Upon arrival, eggs were hatched in 120-l fibreglass incubators at 21 °C. After hatching on February 13, 2004, fish larvae were stocked in three 600-l fibreglass rearing tanks at 60 fish/l. Four feeding regimes included the start of initial feeding on 3 DAH as normal feeding (NF), delayed first feeding until 5 DAH (DF5), a single period of food deprivation from 12 to 15 DAH (DF15), and another single period of food deprivation from 33 to 36 DAH (DF36). In the food deprivation treatments, fish were randomly selected from three normal feeding tanks and moved to three 20-l replicated enclosures which were suspended in each normal feeding tank. Each enclosure was surrounded by 60 (for DF5) and 100 (for DF15 and DF36) μm nylon meshes which allowed free water exchange between the enclosure and the surrounding water in the normal feeding tank, but no food items could enter the enclosure during the period of deprivation trial. All rearing tanks were supplied with filtered seawater through a 5-μm filter in a flow-through system with a water exchange rate of 1.2 l/min at the beginning and increased to 3.0 l/min at the end of this experiment, to wash out all food residues by overnight

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