Ontogenetic changes in schooling behavior and visual sensitivity during larval and juvenile stages in Pacific bluefin tuna, *Thunnus orientalis*

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**Abstract**: Behavioral traits under light and dark conditions were evaluated in larval and juvenile stage Pacific bluefin tuna (PBF), *Thunnus orientalis*. Light/dark conditions affected behavior of PBF throughout the growth stages examined in the present study (from 20 to 55 days post hatching). PBF displayed schooling behavior under light conditions from 27 days post hatching, while they did not display well-coordinated schooling behavior under dark conditions at any point during the present study. Under dark conditions, PBF swam slower than they did under light conditions. From 20 to 29 days post hatching, PBF demonstrated particularly slower swimming behaviors under dark conditions. Results of the present study suggested that both larval and juvenile PBF swam actively under light conditions, and that they formed coordinated schools from 27 days post hatching when exposed to an environment with sufficient visibility.

**Key words**: early life stage, lighting condition, Pacific bluefin tuna, schooling behavior, swimming speed

**Introduction**

Pacific bluefin tuna (PBF), *Thunnus orientalis*, migrate within a wide range, from the western to eastern Pacific, and dive to depths of 120 m below the water surface. These behaviors are made possible by their large body size, strong swimming ability, and physiological traits such as the thermal regulation and visual sensibilities. Various histological and behavioral studies that examined tuna behavior in relation to vision have demonstrated that PBF possess a well-developed vision, which is considered to be their predominant sensory system (Kawamura et al., 2003). Since vision is the predominant sensory system in PBF, their behavior is strongly affected by both light intensity and the differences between light conditions during day and night (Torisawa et al., 2007; Fukuda et al., 2010; Kitagawa et al., 2004). Therefore, developing a greater comprehension of the relationship between behavioral traits and vision in PBF could improve our understanding of how PBF select the environments that they inhabit.

PBF, as well as other pelagic fishes, exhibit typical schooling behavior throughout the majority of their lives (Fukuda *et al.*, 2010). Since forming a polarized school requires precise recognition of the motions of neighboring fish, previous studies have evaluated sensory perception of fishes using variables associated with schooling (Miyazaki *et al.*, 2000, Torisawa *et al.*, 2011). In the present study, behavioral traits associated with schooling in larval and juvenile PBF of different ages were evaluated under light and dark conditions. Schooling variables were compared among different age groups at different light intensities in order to investigate the development of these behaviors as well as the effect of light and dark conditions during early life stages in PBF.
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Materials and Methods

Larval and juvenile PBF used in behavioral experiments were obtained from the Oshima station of the Fisheries Laboratory at Kinki University. The behaviors of 10 fish of different ages (20, 23, 25, 27, 29, 31, 33, 36, 40, and 55 days post hatching (dph)) were examined. In order to regulate conditions in behavioral experiments, such as lighting and water temperature, experiments were conducted in circular tanks (diameter: 0.35 m for 20-27 dph fish, 0.66 m for 29-31 dph fish, 0.90 m for 33 dph fish, and 1.86 m for 36-55 dph fish) situated in a dark room. Illumination was achieved using a halogen light, which provided both dark (<0.01 lx) and light (300 lx) conditions. A digital video camera placed above experimental tanks was used to record the behavior of ten fish for 20 minutes under each experimental condition. Night recording functions of the digital video camera and infrared light-emitting diodes (LEDs) were used to obtain images under dark conditions. During video recording, water exchange and aeration were stopped to prevent stimulation due to water movement. Because the three-dimensional observations would have been difficult owing to the small size and transparent body of the larvae especially in the dark conditions, a shallow water depth to restrict their swimming behavior into the two-dimensional space was maintained.

Using the video images obtained, movements of the 10 individuals examined were digitized in order to obtain time series coordinate data at 0.1 s intervals for periods of 2 min. Behavior of each fish was evaluated using 3 indices: swimming speed, nearest neighbor index (INN) and separation swimming index (ISS). INN has been established as an index of compactness of a fish group (Fukuda et al., 2010) and represents a proportion of nearest neighbor distance (DNN) to expected DNN when fish are distributed randomly within tanks. ISS is expected to be less than one when each individual is positioned close to neighboring fish. ISS is an index of parallel swimming in a fish group (Nakayama et al., 2004). INN was calculated at 2.0 s intervals for all individuals in the tank (n=600 in each experimental condition). In order to calculate ISS, swimming vectors of an individual and its nearest neighbor were estimated over 1.0 s. Distances between paired swimming vectors were then divided by mean absolute values of the two vectors. ISS ranges from zero, when each individual is swimming exactly parallel to its neighbor, to two, when each individual is swimming in the direction opposite that of its neighbor. A mean ISS value when the angle between swimming vectors is between 0 and 180° is used as an expected value (1.27) when each individual swims in a random direction. ISS of 10 individuals were calculated at 10 s intervals for all individuals in the tank (n=120 in each experimental condition).

Swimming speeds (mm s\textsuperscript{-1}) of 10 individuals were measured over 0.3 s at 2.0 s intervals (n=600 in each experimental condition) and scaled by their body length (BL s\textsuperscript{-1}). The detailed information about the calculation of ISS and INN were described in Nakayama et al. (2004) and Fukuda et al. (2010), respectively.

Results

PBF used in the present study ranged from 9.5 mm (20 dph) to 146 mm (55 dph) in average body length (BL). This growth was similar to that reported by Miyashita et al. (2001). The metamorphic change from the larval to juvenile stage was considered to complete at 25 dph (22.7 mm BL). Swimming speed of juveniles increased rapidly from 25 to 40 dph at a rate that exceeded that of body growth (body length: 26.2 to 80.1 mm, swimming speed: 34.4 to 270.7 mm

![Fig. 1. Changes in mean swimming speed during the experimental period. Vertical bars indicate standard deviations. White and black circles indicate swimming speed under light and dark conditions, respectively.](image-url)
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swimming fish (*P* <0.05, *n* = 120; Steel test), and both of the *I*<sub>NN</sub> and *I*<sub>SS</sub> decreased from 25 dph onwards (Fig. 3, Fig. 4). These results indicated that the ontogeny of school formation occurred around 27 dph in PBF; however, PBF did not demonstrate parallel swimming or cohesive aggregation under dark conditions even after 27 dph.

**Discussion**

Metamorphosis from the larval stage to the juvenile stage was completed at around 25 dph in experimental fish. After this point, fish began forming schools, which indicated that timing of the onset of schooling was synchronized with the completion of metamorphosis from larvae to juveniles. These results were consistent with those of

![Fig. 2. Relative frequency distributions of swimming speed scaled by body length of fish (*n* = 600 in each case). White and black bars indicate light (300 lx) and dark (0.01 lx) conditions, respectively.](image)
previous studies, which demonstrated ontogenetic changes in schooling behavior in other species (Masuda & Tsukamoto, 1998; Masuda et al., 2003; Nakayama et al., 2003; 2007). Fish examined in the present study did not form well-coordinated schools at 55 dph under dark conditions. This indicated that fish depended on their vision to perceive the motion of neighbouring fish when schooling.

Larval and early juvenile stage PBF did not actively swim under dark conditions. Takashi et al. (2006) demonstrated that PBF larvae expanded their swim bladder at night, a behavior that decreased their relatively high body density. It was likely that PBF larvae and early juveniles examined in the present study also expanded their swim bladders under dark conditions instead of actively swimming in order to avoid sedimentation caused by their negative buoyancy.

References


Nakayama S., Masuda R., Shoji J., Takeuchi T., and...


