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Measurement of the Swimming Behavior of a Deep-water Fish, the Splendid Alfonsino (*Beryx splendens*), in Captivity using Micro Data Loggers

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ABSTRACT

The splendid alfonsino (*Beryx splendens*) is a deep-water, cosmopolitan fish species that is distributed around seamounts in all tropical and temperate seas, with the exception of the northeastern Pacific Ocean. Micro data loggers were attached to two captive splendid alfonsinos and data on depth, temperature and acceleration (two axes) were recorded over three continuous days. Body tilt angles were extracted from surging acceleration. Pectoral and caudal beat frequencies were determined from swaying acceleration. Both individuals descended to deeper water around sunrise. The first individual always occupied the deeper water, but the second individual inhabited the upper water. This is an indication of space partitioning. Pectoral Beat Frequency (PBF) had higher values than Caudal Beat Frequency (CBF) and was 1.45 times higher in the first individual and two times higher in the second individual. There was no difference in average body tilt angles between day and night in the first individual and values ranged from 20 to 40°. Average body tilt angles varied less in the second individual and averaged approximately 9° during the day and 0° during the night. Both the PBF and CBF were higher in the first individual, presumably to support the high tilt angles. This is the first time that PBF was extracted from the acceleration profile and separated from the CBF. We attributed the positive tilting behavior to the feeding behavior of the splendid alfonsino. This approves the possibility to attach the data logger to the deepwater fish splendid alfonsino and release to the open ocean.

Key words: Acceleration data logger, splendid alfonsino, swimming behavior, body tilt angle, pectoral and caudal beat frequencies

INTRODUCTION

The splendid alfonsino (*Beryx splendens*) is a commercially important fish species in Japan and has a worldwide distribution in temperate and tropical waters, with the exception of the Northeastern Pacific Ocean. They live near seamounts at depths ranging from 25 to 1300 m

(Busakhin, 1982; Morato and Pauly, 2004). They inhabit the flat summits and slope areas of seamounts and their habitat extends to outer continental shelves and slopes. Seamount fishes, including the splendid alfonsino, are highly vulnerable to exploitation compared to other fish groups because of their life history and ecological characteristics of which late maturity, slow growth and low mortality rates (Morato and Pauly, 2004).

Moreover, species that display social aggregation behaviors, such as shoaling, schooling (Pitcher and Parrish, 1993) or shoal spawning, may experience higher vulnerability to exploitation because of increased catchability (Sadovy and Domeier, 2005). This vulnerability can lead to the hyperstability of catch rates (Pitcher, 1995, 1997; Hilborn and Walters, 1992; Walters, 2003) and the possible disruption of group spawning behavior by fishing (Johannes, 1998; Sala *et al.*, 2001; Sadovy and Domeier, 2005). Furthermore, the recent decrease in landings of this species raised concerns about the depletion of its resources (Kuboshima, 1999). For these reasons, proper monitoring and management of the exploitation of splendid alfonsinos should be a priority to conserve and sustain this valuable resource. Due to their steep slopes and rugged ground, seamounts are difficult to sample using conventional methods such as trawling or bottom trawling. Acoustic methods such as an echo sounder represent an alternative approach (Tanoue *et al.*, 2008).

Research on fish behavior is important in the context of biomass estimation of fish stocks using hydroacoustic methods (Huse and Korneliussen, 1995; Orlowski, 1996; Simmonds, 1996). To estimate fish biomass, echo energy reflected from fish schools must be converted into abundance estimates and to achieve this aim, the acoustic Target Strength (TS) of the insonified fish must be measured. Among the many factors affecting TS estimation, tilt angle is shown to have a major influence on TS (Foote, 1980; Blaxter and Batty, 1990; Horne and Jech, 1999) followed by frequency, fish length and depth (Hazen and Horne, 2003).

Because the splendid alfonsino displays aggregation behavior, the nearest neighbor distances are limited (Pitcher and Parrish, 1993). This diminishes the range of movement for each fish in the mean and causes a change in the orientation distribution (Foote, 1980). Moreover, Vinnichenko (1997) suggested that splendid alfonsinos make strong diurnal vertical migrations in the water column. If this hypothesis is true, this behavior will have a major effect on the average body tilt angle. This will affect the acoustic TS of splendid alfonsinos and will thereby influence the resultant fish biomass estimates from acoustic methods. Previously, traditional tag-recapture studies have long been used to examine movement of marine animals and provide an understanding of population dynamics. For example, Sugiura *et al.* (1987) and Sugiura (1990) studied the behavior of splendid alfonsinos using conventional tags. However, the measurement of the fine-scale movements of fish, including the body tilt angle of fishes, was not possible until the recent development of the acceleration data logger that can measure fish orientation accurately *in situ*.

Recent advances in electronic miniaturization, digital information processing and data storage have provided new insights in the behavior of species that can only be studied indirectly through the use of technology (Davis, 2008). Biologging has revolutionized the study of animal behavior and has a promising future for fish behavior studies. Until recently, most biologging studies have been conducted on large marine animals such as whales, seals and seabirds. Few studies have been conducted on fishes. Examples of fish species that have been studied include chum salmon (Kudo *et al.*, 2007; Tanaka *et al.*, 1998, 2000, 2001), Japanese flounder (Kawabe *et al.*, 2003a, b, 2004), Chinese sturgeon (Watanabe *et al.*, 2008) and sunfish (Watanabe and Sato, 2008). However, there are not any studies on the behavior of deep-water fishes using data loggers, because of the problems inherent in their small sizes and the stress they encounter when they are brought to surface. This is, to our knowledge, the first attempt to study the behavior of a deep-water fish using data-logging technology. Since the advent of acceleration data loggers, few studies have utilized these instruments for the measurement of body tilt angle of fishes. Tanaka *et al.* (2001) conducted the first study on chum salmon. This study was followed by a study on Japanese flounder by Kawabe *et al.* (2004) and a recent study conducted by Tanoue (2009) on threeline grunt in cages where he reported a significant difference in body tilt angles between day and night.

The present study describes the swimming behavior of splendid alfonsinos in captivity using an acceleration data logger. The swimming depth, fin beat activity and body angle were studied and the results are discussed with respect to the relationships between these parameters and the swimming behavior of splendid alfonsinos. Moreover, we report here the body tilt angle distribution, PBF and CBF of splendid alfonsinos and discuss the results in respect to the other fish behaviors. This is the first successful experiment conducted on fishes to measure the frequency of PBF.

MATERIALS AND METHODS

Twenty three individuals were caught using hook and line by the R/V *Enoshima-Maru* in Sagami Bay, Japan, on March 28, 2009. Fish were transported using a 1 ton container to the laboratory of the National Research Institute of Fisheries Science, Fisheries Research Agency, Kanagawa, Japan where the experiment was conducted. Then, two out of the twenty three individuals were chosen to conduct the experiment taking into account that they were perfectly healthy. We used an experimental tank that was 100 cm long, 80 cm wide and 100 cm deep. Water temperature was maintained at 13°C and the light environment was kept dark to maintain natural habitat conditions in the tank. The tank was provided with aeration throughout the period of the experiment. The fish were allowed four days to acclimate to the new environment before the experiment was conducted. Tank was ensured to be stress free environment for splendid alfonsino according to the requirements of animal welfare of the Farm Animal Welfare Council (FAWC) as described by Sejian *et al.* (2011). The total length and weight of each fish were measured and the data logger was attached to a float of copolymer foam (Nichiyu Giken Kogyo Co., Saitama, Japan) to achieve neutral buoyancy. The data logger was sutured with plastic cables to the body of each fish in front of the dorsal fin. The experiment was conducted from 2 April until 5 April 2009 and the fish were left alive in the tank for another one week before the data loggers were detached from the fish. The behavior of the splendid alfonsinos was monitored using a 12-bit resolution, 128 megabytes of memory, four-channel UWE-D2GT logger (45 mm in length, 12 mm in diameter and weighing 10 g in air; Little Leonardo, Tokyo, Japan). The logger recorded the depth, temperature and two axes of acceleration. Depth and temperature (fixed at 13°C) were recorded at a frequency of 1 Hz and acceleration data were measured at a frequency of 16 Hz. Acceleration data were recorded along the longitudinal (surging) and right-left (swaying) axes of the fish (Yoda *et al.*, 2001). Fish were observed for 1 h using a video camera. Video recordings allowed us to compare visual observations and data logger records of swimming behavior and to calculate PBF and CBF. Data were downloaded from the accelerometers to a computer and analyzed using Igor Pro (ver. 6.03J, Wave-Metrics) and Igor Filtering Design Laboratory (IFDL Ver. 4, WaveMetrics). The center of mass (n = 7) was measured as described by Yanase and Arimoto (2007) and x-ray images (n = 7) of the whole fish body of anesthetized individuals were taken for both instrumented (equipped with data loggers, n = 2) and uninstrumented (not equipped with data loggers, n = 5) individuals. Then, all individuals were dissected to examine the physical condition of the swim bladder.

Swaying accelerations often contained low frequency variations. These variations were separated using a high-pass filter. Surging acceleration was filtered using a low-pass filter (IFDL Ver. 4, WaveMetrics) to remove the acceleration of the movement following Watanabe *et al.* (2004). The acceleration data along the longitudinal body axis were then converted to sine values and finally to degrees. Swaying acceleration contained two types of movements, high frequency and low frequency, which corresponded to PBF and CBF, respectively. Along with data logger measurements, we captured a movie of the fish swimming to calculate the average values of PBF and CBF from the visual observations. Using these values, swaying acceleration was subsequently subdivided into two components (PBF and CBF) from data logger measurements.

RESULTS

Visual observation with video-imaging: The fish were observed by a video camera for one hour to compare between visual observations and data logger records of swimming activities. We could find no difference between instrumented and uninstrumented individuals. We calculated both the PBF and CBF from the recorded movie and the differences between them were significant. The PBF was higher than the CBF in both individuals. The bigger individual always occupied the upper part of the tank and the other individual occupied the lower part of the tank with occasional rapid visits to the surface layer.

Alfonsino has been recognized to use mainly the pectoral fins for swimming, moving upwards and especially for posture stablization. We have also noticed from the movie that the bottom individual had a tilting behavior at low swimming speeds associated with high rate of PBF. The fish that occupied near the bottom displayed much higher positive tilt angles, especially when swimming at low speeds. As speed decreases, tilt angle increases and the highest tilt angle (around 50°) reached when hovering at zero swimming speed. Moreover, if the near surface fish at any time descended to near the bottom, they displayed positive body tilt angles generally increasing as speed decreases.

Behavior of splendid alfonsino using the accelerometer: Depth, temperature and acceleration data were recorded for 72 h for each individual. One hour average time series data for both individuals showed that the behavior of the splendid alfonsinos were stable during the last two days of deployment. Therefore, we used only the data from days two and three for subsequent analyses of depth, fin beat frequencies and body tilt angles.

The swimming depth, PBF, CBF and body angle data for both individuals are shown in Fig. 1a and b. Both individuals descended to deeper water around sunrise and they did not show any specific behavior during sunset (Fig. 2). We calculated the average swimming depth, PBF, CBF and body tilt angles at day and night to compare changes in the behavior between day and night. We found that the first individual always occupied deeper water compared to that occupied by the second individual. Both fish always kept a vertical distance between each other. The two individuals occupied an average depth of 0.51±0.11 and 0.25±0.08 m for the first individual and the second individual, respectively (Fig. 3). The upper individual and the lower individual had a fork length of 23.5 and 24.5 cm respectively and they had respectively a total weight of 307.9 and 371.1 g.

The PBF and CBF from the recorded visual data were used to extract their values from the swaying acceleration. Figure 4 shows a sample of the resulting PBF and CBF. It shows that the PBF is higher than the CBF (e.g., the averages of PBF and CBF of the first individual calculated from visual observation were 2.39±0.39 and 1.81±0.14 b/s, respectively). Therefore, we were able to separate the PBF and CBF from the swaying acceleration. The PBF and CBF of the first

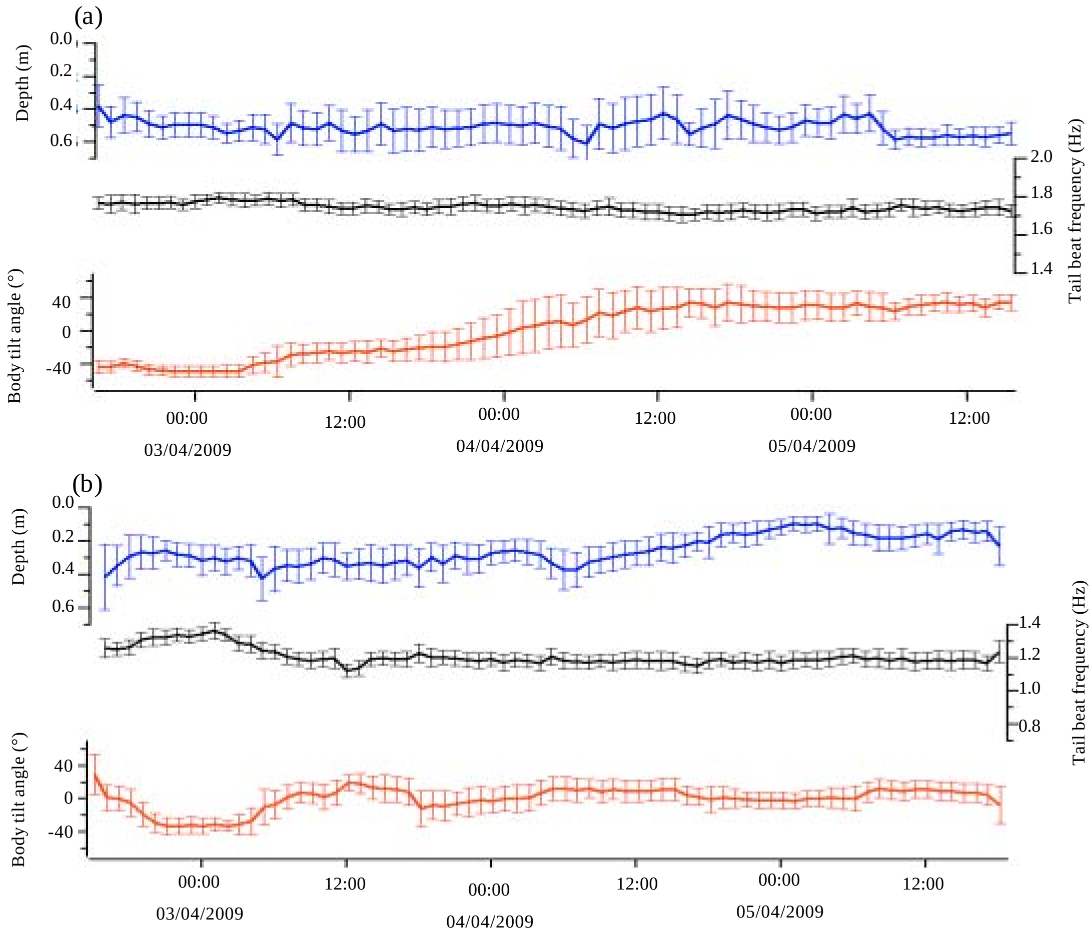

Fig. 1: One-hour average time series data of depth, body tilt angle and tail beat frequency of the first individual (a) and the second individual (b)

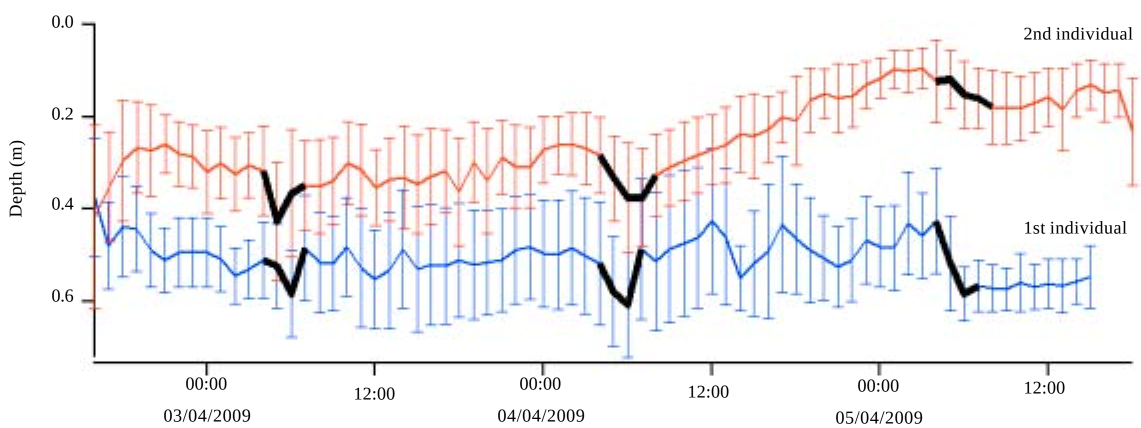

Fig. 2: One hour average time series data of depths occupied by the two individuals. Thick areas of the wave corresponds to sunrise time

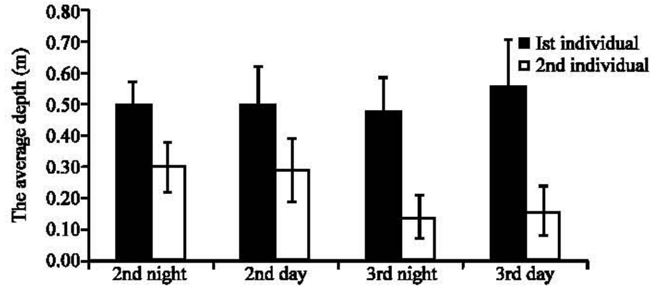

Fig. 3: The average depths occupied by the two individuals. The first individual always occupied deeper waters compared to the second individual throughout the experiment. Vertical bars represent standard deviation

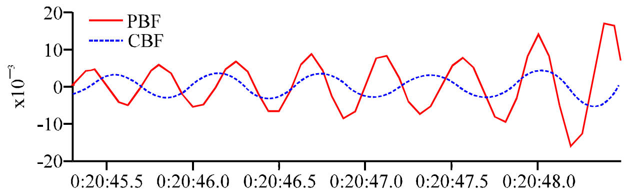

Fig. 4: A sample of the resulting PBF and CBF from 0:20:45:0 to 0:20:48:5 on 4 April 2009. The pectoral beats had higher frequencies than the caudal beats

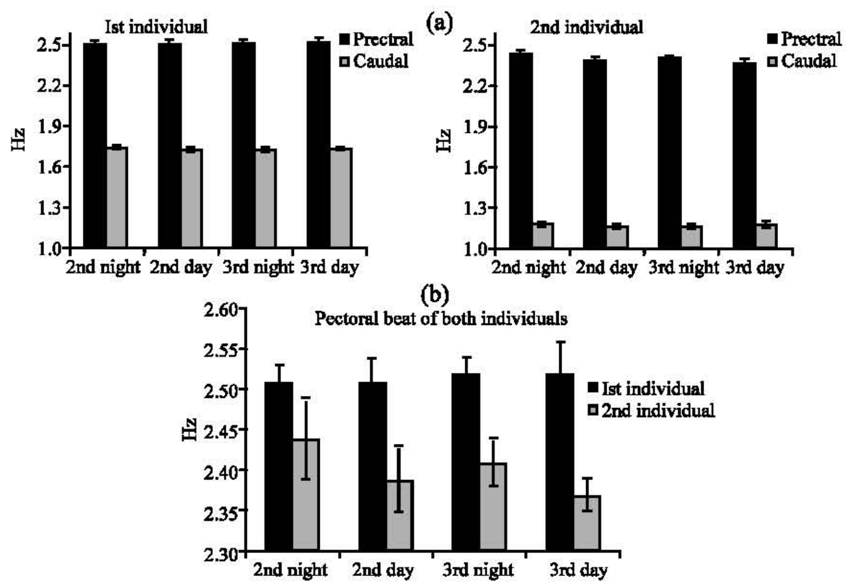

Fig. 5: (a) The average frequency of PBF and CBF of both individuals. The PBF was much higher than the CBF for both individuals and the CBF was much higher in the first individual. (b) PBF of both individuals. PBF was higher in the first individual. Vertical bars represent standard deviation

individual (near the bottom) had, respectively an average of 2.52±0.03 and 1.74±0.01 beats per second (b/s) and they had, respectively an average of 2.42±0.03 and 1.19±0.02 b/s in the second

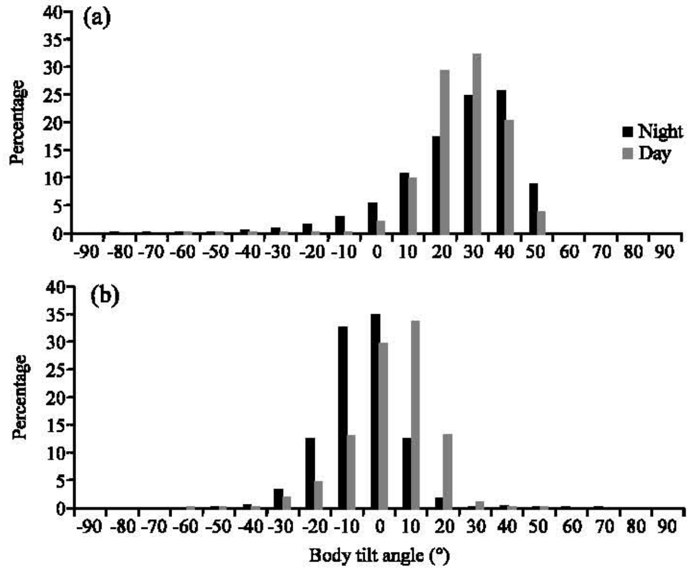

Fig. 6: Frequency distributions of body tilt angles of both individuals over the entire recording period. (a) 1st individual and (b) 2nd individual

individual (near the surface). The PBF was 1.45 times higher than the CBF in the first individual and it was two times higher than the CBF in the second individual. Both the PBF and CBF were higher in the first individual which occupied the water near the bottom of the tank. The CBF of the first individual was 1.47 times higher than that of the second individual (Fig. 5a). The PBF of the first individual was higher than that of the second individual (Fig. 5b).

The first individual showed positive body tilt angles during both daytime and nighttime. The second individual had a normal distribution of body tilt angles, with more positive angles during the daytime and more negative angles (an average near zero) during the nighttime. Body tilt angles did not differ between day and night in the first individual and ranged from 20 to 40° (The average at day 18.7°, at night 23.3°). The average body tilt angles changed less in the second individual, with values around 9°±12.45 during the daytime and close to 0±14.1 during the nighttime (t-test, $p<0.01$). The distribution of body tilt angles for both individuals is shown in Fig. 6a and b.

Center of mass and X-ray imaging: The center of mass (n = 7) had a posterior upper position to the base of the pectoral fins. X-ray images showed that the splendid alfonsino has a tilted swim bladder. X-ray images also showed that the swim bladder of the splendid alfonsino is tilted with an average angle of 10±6° (n = 7) between the longitudinal axis of the swim bladder and body axis (Fig. 7a-c). The dissected fish showed that the swim bladder of both instrumented and uninstrumented individuals did not burst.

DISCUSSION

We collected data on the behavior of the splendid alfonsino for three continuous days and the fish remained healthy until we detached the data loggers one week after the end of the experiment. That proved the possibility to attach the data logger to the deep-water fish alfonsino and release to the open ocean. The fish movements were stable during the last two days as an indication of the

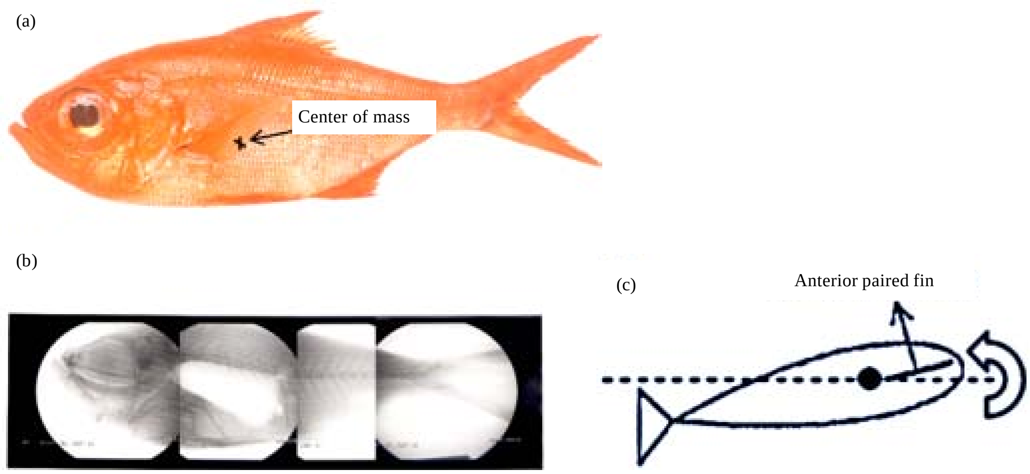

Fig. 7: (a) Location of the center of mass. (b) X-ray image of a splendid alphonsino (*Beryx splendens*) showing that the swim bladder is tilted. (c) The pectoral fins anterior to the center of mass function to increase the angle of the pectoral fins and hence the lift generated by these fins and this ultimately makes the fish body tilt upward (Webb, 2005)

acclimation to the tank environment. Therefore, we used this data for subsequent analysis of depth, fin beat frequencies and body tilt angle distribution. The two individuals occupied different depths, with the first individual mostly occupying the upper part and the second individual occupying the lower part of the tank. Begon *et al.* (1996) showed that the intensity of intraspecific competition experienced by an individual is not determined by the density of the population as a whole, but by the extent to which it is crowded or inhibited by its immediate neighbors. Accordingly, it appears that splendid alfonsinos always keep a spatial distance between each other as an indication of space partitioning. In addition, the upper waters seemed to be favored, because the individual on the surface could search for food at a broader scale on horizontal, downward and upward planes. In contrast, the individual on the bottom could only access the food in the horizontal and upward planes. Moreover, the PBF and CBF were less in near the surface individual than in the bottom individual, indicating less energy consumption and this add an advantage to staying in the upper layer. The bigger individual always occupied the better position at the upper part of the tank. This finding was supported by the visual observations from the video camera and was clear from the depth data, since the bottom individual always tried to ascend to the surface water but the space was already occupied by the other individual.

The two individuals always descended around sunrise in a behavior similar to that found in nature, though they did not show any specific behaviors around sunset. Some previous studies have suggested that splendid alfonsinos ascend to surface waters at night to feed on the small pelagic fish and occupy depths around seamounts during the daytime (Galaktionov, 1984; Horn and Massey, 1989; Vinnichenko, 1997). Komatsu *et al.* (2002) showed using acoustic data that splendid alfonsinos occupy depths above the bottom at nighttime and suggested that splendid alfonsinos occupy habitats near the bottom during the daytime (unpublished). Fishermen usually catch splendid alfonsino above the bottom at nighttime using hooks and vertical lines and near the bottom during daytime using horizontal lines.

We noticed from the recorded movie that the splendid alfonsinos use the pectoral fins for swimming. The PBF of the individual that stayed near the bottom was 1.45 times higher than its

CBF and the ratio was two times higher for the fish that stayed near the surface. These findings indicate that the splendid alfonsinos mainly use the pectoral fins for swimming, especially for moving upward, as confirmed by observations from the video camera. Most fishes use their pectoral fins in some way for steady swimming, turning, braking, or balance. For many species, the labriform mode (pectoral fin propulsion) is the primary mode of swimming (Westneat and Walker, 1997). Oscillatory pectoral fins are routinely used by many fish as auxiliary propulsors and they can also provide adequate thrust to be used as the sole means of locomotion, at generally low speeds (below 3 BL/s) (Sfakiotakis *et al.*, 1999).

Because the center of mass of splendid alfonsinos has an anterior position and the swim bladder is tilted (Fig. 7a, b), the fish normally positioned head-down when they are immobile. Threeline grunt, had a tilted swim bladder with an angle of 19.0±1.4°, displayed head-down orientation during immobile periods in which the longitudinal axis of the swim bladder become horizontal (Komatsu *et al.*, 2010). However, the splendid alfonsino feed on prey in the upper layer that are located in their field of vision (which is confined to the horizontal and upward planes) and they do not feed on benthic prey (Kuboshima *et al.*, 1998). Therefore, for the fish to feed on the prey in the upper layer, they have to tilt their bodies upward. When the pectoral paired fins have an anterior position (to the center of mass), as is the case in splendid alfonsino, they are used by the fish to generate lift and increase the pitching (Webb, 2005) (Fig. 7c). Thus, the higher body tilt angle recorded for the bottom individual was attributed to the high rate of PBF.

The fish that occupied near the bottom exhibited head-up swimming and steeper positive body tilt angles during both the daytime and nighttime, especially at low swimming speeds. It reached maximum body tilt angles when it was hovering at zero swimming speed. This finding is supported by the high values of the PBF and CBF. Body tilting by fish is a general phenomenon associated with slow swimming and has been proposed to be a behavioral mechanism to increase stability control at low swimming speeds (Webb, 1993). In addition, tilting has been proposed to increase the total area generating lift by using the body as a hydrofoil at low swimming speeds when lift from the pectoral fins is presumably insufficient (He and Wardle, 1986; Wilga and Lauder, 2000). As speed decreases, fishes swim at increasing angles (higher tilt) (He and Wardle, 1986; Webb, 1993; Ferry and Lauder, 1996; Wilga and Lauder, 1999, 2000). This behavioral pattern of posture stabilization allows the fish to maintain a chosen swimming depth (He and Wardle, 1986) and minimizes energy costs by orientating the body to minimize drag (Weihs, 1993).

Tilting increases resistance and thereby increases tailbeat frequencies at low swimming speeds (Webb, 2002). This was apparent by the splendid alfonsino that stayed close to the bottom and displayed higher PBF and CBF values, presumably to provide support for the high head-up posture. With higher tilt, the PBF increased (as confirmed by the visual observations). Moreover, when hovering at zero swimming speed, the splendid alfonsino that stayed close to the bottom displayed higher PBF and CBF values. When hovering at zero swimming speed, fishes use powered forces alone, such as pectoral and caudal beats, for stability (Blake, 1979).

The fish that stayed near the surface had smaller positive body tilt angles during the daytime because splendid alfonsinos in nature usually live near the bottom during the daytime. This individual even stayed away from the bottom during the experiment, but tried to maintain the same behavior of splendid alfonsinos in nature, which is to be directed upward during daytime and have positive body tilt angles. In addition, the fish during the nighttime are usually active in searching for food and are expected to have a range of body tilt angles. Moreover, if the surface fish in our experiment descended near the bottom, it had a positive body tilt angle. This is the usual behavior of splendid alfonsinos that occupy habitats near the bottom.

CONCLUSION

The acceleration data logger has proved useful for measuring the behavior of the splendid alfonsinos and upon the attachment of the data logger to two splendid alfonsinos we could find no effect on their behavior after the first day of the experiment. That proved the possibility to attach the data logger to the deep-water fish splendid alfonsino and release to the open ocean. Moreover, the data logger is a useful tool to measure fin beat frequencies and body tilt angle. Since the pectoral and caudal beats of splendid alfonsinos have different frequencies, with the pectoral fin having higher frequencies when compared to the caudal fin, it was easy to extract them from the swaying acceleration. This is the first time that the PBF was extracted from the acceleration profile and separated from the CBF.

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