Stomach content of ichthyofauna present at the vicinity of Cobia fish culture cages in Bahía de Cochinos, Cuba

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ABSTRACT

Stomach content analysis is one of the most efficient methods to identify dietary components in the marine organisms and to infer feeding behaviors. This study was aimed to determine the natural diet composition of ichthyofauna present at the vicinity of Cobia fish culture cages in Bahía de Cochinos, and also to infer changes in the trophic spectrum of this ichthyofauna. The stomach contents of 103 fish individuals were analyzed after being collected between May and June 2015 in the morning and afternoon. The frequency of occurrence index (FO), gravimetric analysis (G) and relative importance index (RII) were calculated. Fish species with the highest representation in the collections were yellowtail snapper (*Ocyurus chrysurus*, 68.9 %) and blue runner (*Caranx crysos*, 22.3 %). There were 16 feed items identified belonging to five groups: fish, crustaceans, mollusks, echinoderms, and vegetable remains. In both periods (morning and afternoon) preferential preys were fish (FO=57.1 % and 65.7 %, respectively). Furthermore, feed remains were found and were classified as a secondary item based on their occurrence. However, according to G and RII indexes, it was the most important item according to weight and the unique with at least secondary relative importance. Feed ingestion moves fish trophic spectrum mainly at feeding times in the cages and, allows a decrease of organic matter accumulation under cages and facilitates its suspension and transport to other areas in the vicinity of the cages.

Key words: aquaculture, Rachycentron canadum, aquatic organisms.

Contenido estomacal de la ictiofauna presente en la cercanía de jaulas de cultivo de peces Cobia en Bahía de Cochinos, Cuba

RESUMEN

El análisis del contenido estomacal es uno de los métodos más eficientes para identificar componentes dietéticos en organismos marinos e inferir comportamientos alimentarios. El objetivo fue determinar la composición de la dieta natural de la ictiofauna en la cercanía de jaulas de cultivo de peces Cobia en Bahía de Cochinos e inferir el efecto del cultivo en el espectro trófico de esta ictiofauna. Se analizó el contenido estomacal de 103 ejemplares colectados entre mayo y junio del 2015, durante la mañana y la tarde. Se calculó la frecuencia de ocurrencia (FO), análisis gravimétrico (G) y el índice de importancia relativa (IIR). Las especies de peces con mayor representación en las colectas fueron las rabirrubias (*Ocyurus chrysurus*, 68,9 %) y las cojinúas (*Caranx crysos*, 22,3 %). Se identificaron 16 entidades alimentarias pertenecientes a 5 grupos de organismos: peces, crustáceos, moluscos, equinodermos y material vegetal. En ambos períodos, los peces fueron las presas preferenciales (FO=57,1 % y 65,7 %, respectivamente). Se encontraron además restos de pienso, el cual se clasificó como entidad secundaria con base en su ocurrencia. No obstante, los valores de G y IIR mostraron que es el más importante en términos de peso y el único con importancia relativa secundaria. La ingestión de restos de pienso desplazó el espectro trófico de los peces hacia dicha entidad alimentaria, principalmente en los momentos de suministro, disminuyendo la acumulación de materia orgánica debajo de las jaulas y facilitó su resuspensión y transporte hacia otras zonas del ambiente cercano a las jaulas.

Palabras clave: acuicultura, Rachycentron canadum, organismos acuáticos.

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INTRODUCTION

Cage aquaculture has grown rapidly over the last decades and currently undergoing major changes in response to pressures from globalization and increase in global demand for aquatic products (Halwart et al. 2008). The impact of aquaculture on floating cages is small and highly localized. However, seafloor close to such cages is subject to a large deposition of fecal material and unconsumed food (Vita et al. 2002). Increase in organic matter causes physicochemical sediment changes, which causes a reduction in the biological diversity of benthic organisms. However, in some cases they offer new resources to wild fish populations, which allows densities of some species of fish and invertebrates of surrounding communities to increase (Vita et al. 2002).

Food is an essential element of the aquatic environment which influences growth, survival and reproduction of organisms (Brewer and Rabeni 2008). It is also a key factor in the fish abundance and defines the structure of communities limiting the number of species and individuals that can coexist in the same area (Piet *et al.* 1998, Hernández-Aguilar *et al.* 2016).

In most cases, it is difficult to observe fish feeding. Therefore, analysis of the stomach contents becomes one of the most efficient methods to identify the components of the natural diet (Estupiñán-Montaño *et al.* 2018). This test is a valuable tool to infer how energy flows in a trophic network and to determine fish interactions with other species within a community (Krebs 1989, Hernández-Aguilar *et al.* 2016). Moreover, this method allows estimating the organisms' variability in the feeding areas through the description and quantification of dams (Canto-Maza and Vega-Cendejas 2008, Valenzuela-Quiñonez *et al.* 2018).

Nowadays Cuba is realizing various aquaculture activities with different species. One of the most expectative is on the Cobia (*Rachycentron canadum*; Linnaeus 1766), which is now developing for first time. Estimating the Cobia aquaculture impact on natural environment is necessary for a quickly and healthy developing of this activity and to infer the culture effect in the fish trophic spectrum found at the site. The aim of this study was to determine the natural diet composition of ichthyofauna associated farming cages holding Cobias at the Bahía de Cochinos, Cuba.

MATERIALS AND METHODS

Study location

This study was carried out in the Bahía de Cochinos, at 22°12'51" N, 81°11'36" W (Figure 1). At this location two Cobia fish culture cages were placed; each cage was a cylindrical structure (19 m of diameter) and were situated following the consideration of Norwegian specialists (personal communication). The cages bottom was located at 8.0 m and the maximum depth at the sampling site was 18 m. This is first pilot experience in Cuba and began with 1230 Cobias per cage.

Fish catch and sample collection

A sample of 103 stomachs of caught fish around Cobias farming cages, was analyzed. Catches were made in May and June 2015 at morning (between 6 a.m. and 12 m.) and afternoon (from 12 m. to 5 p.m.), through hook and line, using fish flesh as bait. Each specimen was recorded with species name, time of capture, total length (cm) with a precision of 0.1 and total weight (g) with a precision of 0.1. Fish species identification was based on the criteria of Guitart (1974), Humman (1997) and Claro *et al.* (2001).



Figure 1. Location of the sampling area in the Bahía de Cochinos, Cuba.

For extraction of stomach contents, each fish was eviscerated in the field after capture, making a ventral longitudinal cut from the top edge of the opercula to the anus. The stomach was separated from the esophagus and intestine using cuts on the cardias and on the pyloric caeca; then, the stomach was knotted at both ends to prevent loss of content and preserved by freezing for further analysis.

Laboratory analysis

The stomachs were weighed before and after extracting their contents, using a digital technical balance (Kern EMB 600-2; precision of 0.01 g). The stomach contents were analyzed using a stereomicroscope and specialized literature (Clothier 1950; Zúñiga-Romero 2002a, 2002b; Redfern 2013); the items present were identified to the lowest taxonomic level permitted by the food digestion level. Bait fragments were excluded from processing. The following determinations were made from the analysis of the stomach content: repletion index (RI), vacuity coefficient (V), frequency of occurrence index (FO), gravimetric method (G) and the relative importance index (RII). Repletion index was determined according Franco and Bashirullah (1992).

$$RI = \frac{\text{weight of stomach contents (g) x 100}}{\text{fish weight (g)}}$$

Based on this index, the stomachs were classified as full (RI \ge 1), half full (0.5 \le RI \le 1) and empty (RI \le 0.5). Vacuity coefficient was calculated according to the following equation.

$$V = \left[\frac{empty \ stomachs \ (Ev)}{total \ number \ of \ stomachs \ (Et)}\right] \times 100$$

The quantitative analysis was based on the indexes determination frequency of occurrence index (FO) and gravimetric method (G), proposed by Hyslop (1980).

$$FO = \frac{n}{NE} \times 100$$

Where:

n = number of stomachs containing some prey item.

NE = total number of stomachs with food.

$$G(\%) = \frac{p}{PT} \times 100$$

Where:

p = prey weight.

PT = total weight of prey items.

The entities found were grouped as accidental (FO < 10), secondary (10 < FO < 50) and preferential (FO > 50), according the food categories proposed by Franco and Bashirullah (1992). To estimate the importance of each food category in the diet composition, the relative importance index (RII) was calculated using the equation modified by Olaya-Nieto *et al.* (2003).

$$RII = \frac{FO(\%) \times G(\%)}{100}$$

For the classification of RII, the following criteria were used: 0-10 % lower relative importance, 10-40 % secondary relative importance and 40-100 % high relative importance.

Comparisons were also made between two different periods according to capture time: morning (between 6 a.m. and 12 m.); afternoon (from 12 m. to 5 p.m.). To evaluate the differences in FO between the periods, a bifactorial ANOVA was used. G index was determined for items with a greater weight than 0.1 g.

RESULTS AND DISCUSSION

The captured fish belonged to 10 species. Total length of caught fish ranged between 16 and 75 cm with an average weight of 262 g. Fish largest congregations were sighted between 5 and 10 m of depth, near the cages bottom. The species with the highest representation in the captures were the yellowtail snapper (*Ocyurus chrysurus*; Bloch 1791) with occurrence of 68.9 % and blue runner (*Caranx crysos;* Mitchill 1815) with 22.3 % (Table 1). The calculation of RI showed that most of the stomachs were empty (56.3 %), while 7.8 % were classified as half full and the remainder as full (35.9 %). Vacuity coefficient was 21.4 %.

The high proportion of empty stomachs (RI = 56.3 %) may be due to the expulsion of ingested organisms caused by post-capture handling. Regurgitation produced by the contraction of the

20	18

Scientific name	Specimens (n)	Occurrence (%)	Stomachs with food content (n)	Empty stomachs (n)
Ocyurus chrysurus	71	68.9	56	15
Caranx fusus	23	22.3	18	5
Sparisoma viride	2	1.9	2	0
Lutjanus analis	1	1	1	0
Caranx latus	1	1	0	1
Caranx ruber	1	1	1	0
Lutjanus apodus	1	1	1	0
Calamus calamus	1	1	1	0
Rachycentron canadum	1	1	0	1
Dasyatis sayi	1	1	1	0
Total	103	100	81	22

esophageal muscles, have been described as a strategy of defense and escape (Olaya-Nieto *et al.* 2009), but can also occur when fish are rising rapidly or in response to a violent catch. This phenomenon can be stimulated by the use of hooks (used in this study) and gillnets to catch individuals (Rojas 1996). On the other hand, it is usual to find a higher proportion of empty stomachs in carnivorous fish (the most abundant in this study), because they have stomachs with greater volume than herbivores and omnivores (Olaya-Nieto *et al.* 2009).

Sixteen food categories were established from the identified material (both from plant and

animal origin) belonging to the following groups of organisms: fish, crustaceans (crabs, shrimps, amphipods, isopods and copepods), mollusks (gastropods, bivalves and polyplacophorans), echinoderms (brittle stars and sea urchins) and vegetable remains. Further, artificial feed remains provided to Cobia, were found in the stomachs evaluated. This item was named food (F) and together with the remains of crustaceans, shells and plants, formed the group of secondary elements, according to their frequency of appearance. Except for fish remains, a unique preferred item, the other food categories were classified as accidental (Figure 2).



Figure 2. Frequency of occurrence (FO) of food items present in the stomach contents of captured fish species (FR: fish remains, F: feed, CR: crustacean remains, SR: shell remains, B: bait, VR: vegetable remains, I: isopods, MR: mineral remains, OM: organic matter, A: amphipods, O: opercula, CP: chiton plates, CL: crustacean larvae, BSR: brittle stars remains, SUR: sea urchins remains and C:copepods).

The repletion index analysis showed a higher percentage of fish caught on the afternoon, had full stomachs (RI = 41.5%), compared to fish caught on the morning (RI = 16.7%). Conversely, empty stomachs were less numerous on the afternoon (RI=51.2%) than on the morning (RI=62.5%). Similarly, the half full stomachs were less frequent on the afternoon (RI=7.3%) than on the morning (RI=20.8%).

The preferred food item was fish remains in both periods (FO=57.1 % and 65.7 %, respectively). The remaining food items were less important in the fish diet, for the same periods and were considered secondary or accidental (Figure 3). Referring all identified item (Figure 4A) no significant difference were found. Similar results were obtained when comparing item grouped by food categories (Figure 4B).

That could be explain by the fact that feed were almost the unique item found in those stomachs. Most number of fish were caught on the afternoon, around 5:00 p.m., while supplying feed for the Cobias or soon after. The similar frequency of occurrence of food categories in both periods,



Figure 3. Frequency of occurrence (FO) of food categories present in the stomach contents of captured fish species, according to capture time (F: balanced feed, FR: fish remains, CR: crustacean remains, SR: shell remains, MR: mineral remains, VR: vegetable remains, OM: organic matter, SUR: sea urchins remains, CP: chiton plates and BSR: brittle stars remains).



Figure 4. Bifactorial ANOVA for comparison FO between both periods (morning and afternoon), (A): showing all identified items and (B): showing classified food categories (**Pre:** preferential, **Sec:** secondary and **Acc:** accidental).

could mean similar diets. It shows a balance in the consumption of prey between both periods. So, regarding FO, it suggests the same consumption (morning and afternoon) for the fish at the vicinity of Cobia culture cages.

Fragments of gastropod species like *Cerithium litteratum* (Born 1778), *Diacavolinia longirostris* (Blainville 1821) and the bivalve *Tagelus* sp. (Gray 1847) were identified. Vegetable remains were represented by marine plants, mainly *Thalassia testudinum* (Koenig 1805).

The weight determined by gravimetry, in addition to the RII, showed that the balanced feed was the most representative item, also the only food item with secondary relative importance. The other food items were included into the low importance category (Figure 5A and B, respectively).



Figure 5. Gravimetric Index (A) and Relative Importance Index (B) of food items present in the stomach contents (F: balanced feed, FR: fish remains, CR: crustacean remains, OM: organic matter, MR: mineral remains, SUR: sea urchins remains, SR: shell remains, I: isopods and VR: vegetable remains).

This corroborates the reports by Bjordal and Skar (1992) and Valle-Pérez (2005), which indicated that feed not eaten by Cobias is consumed by fish at the vicinity of area. The preferential intake of this item could be caused by its greater palatability compared to natural foods. The feed granules are composed of fishmeal, soybean meal, fish oil, gluten meal, corn, wheat, vitamins and minerals that improve their organoleptic properties (color, smell, taste, texture), and became a most attractive food.

The bromatological composition of extruded feed (according to the manufacturer Skretting, Canada) indicates that it contains between 50 - 55 % of crude protein, 15 - 18 % of crude fat and 1.5 % of crude fiber, which supplies the fish's energy requirements. The main advantages of extruded feeds are: better digestibility of carbohydrates, better utilization of vegetable protein, slower digestion and increased floatability and stability of the granules (Vergara-Martín *et al.* 2005). This last advantage allows fish to capture feed particles with less energy investment than capturing natural preys.

The stomachs containing feed (66.6 %), also had scales in them. Most of these stomachs did not have any other fish remains inside (like fishbones, muscle, fins). This could be because the sampled specimens and Cobias fish ate feed at the same time, thus ingesting the released scales from animals in captivity.

Feed consumption by fish at the vicinity farming cages prevents the accumulation of food that would drop to the bottom and its negative effect on benthic communities and sediments. The effects on the environment due to the discharge of organic materials from aquaculture facilities are softened by wild ichtyofauna (Vita *et al.* 2002). This aspect is also favored by other factors such as speed and direction of marine currents, water temperature, depth of the site and the physical and chemical properties of waste particles (Buschmann 2001).

The attraction of fish to the floating cages is due to several factors, like increased availability of food resources and improved habitat structural diversity, among other (Gooding and Magnuson 1967, Lopeztegui-Castillo *et al.* 2018). In addition to the availability of artificial feed, a lot of invertebrates associated with nets, tubes, ropes, buoys and other devices of the floating structure, are preys for some fish.

Furthermore, culture cages also increase the refuge availability, contributing to the aggregating effects on the fish species in the surroundings. These structures are often used by some pelagic and demersal fish as recruitment areas (Deudero *et al.* 1999, Lopeztegui-Castillo *et al.* 2018). Fish aggregations around the cages primarily include small specimens that favor the arrival of bigger predatory fish species, increasing the biomass of the different levels in the food web.

The diet of the yellowtail snapper (the most abundant species) included a wide variety of prey items. According to Claro and Lindeman (2008), its high richness diet is a particular characteristic of the species trophic biology. This may be a consequence of its pursuit behavior for prey, since this snapper, in addition to feeding at night, like other snapper species, does not restrict its feeding activity to the benthic zone and also feeds through the column of water and even on the surface (Sierra 1996). These characteristics promote *O. chrysurus* as a typical species that lives at the vicinity of floating aquaculture cages.

CONCLUSIONS

Most abundant fish at the vicinity of *Rachycentron canadum* culture cages were the yellowtail snapper (*Ocyurus chrysurus*) and the blue runner (*Caranx fusus*), which respectively represented the 68.9 % and 22.3 % of a total 103 captured fish.

The natural diet of the ichthyofauna at the vicinity of *Rachycentron canadum* culture cages in Bahía de Cochinos, is composed of 16 food items belonging to the following groups: fish, crustaceans (crabs, shrimp, amphipods, isopods and copepods), mollusks (gastropods, bivalves and polyplacophorans), echinoderms (brittle stars and sea urchins) and plant remains. Only fish remains were classified as preferential item (high frequency of occurrence), which confirms that captured individuals were mainly ichthyophagi.

The feed constituted, along with crustaceans, shells and plant remains, the group of secondary items based on the frequency of occurrence. However, feed was the most significant item in terms of weight (gravimetry) and relative important index (RII), evidencing their representativeness in the diet of analyzed fish.

Most fish at the vicinity of the culture cages are shifting their trophic spectrum toward feed item,

which does not imply the absence of natural food items in their diet.

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