Land-Based Marine Aquaculture: Pilot Study on Cobia *Rachycentron canadum* in Recirculating Aquaculture Systems in Trinidad and Tobago

Acuicultura Marina en Tierra: Estudio Piloto Sobre las Cobias *Rachycentron canadum* en los Sistemas de Recirculación Acuícola en Trinidad y Tobago

Aquaculture Marine sur Terre Ferme: Étude Pilote sur le Cobia *Rachycentron canadum* dans les Systèmes d'Aquaculture en Parcs Clos à Trinité-et-Tobago

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ABSTRACT

A pilot study was conducted on cobia *Rachycentron canadum* to determine operating parameters for culture, technical feasibility, and associated challenges in a marine recirculating aquaculture system (MRAS) under local conditions. Juvenile cobia 2.8 ± 0.9 g were imported from Florida, USA and quarantined in three 2800 L circular polyethylene tanks attached to a 680 L mechanical filter filled with 0.34 m^3 Kaldness® media, a 680 L sump, two 110W ultraviolet sterilizers and moving bed bioreactors (MBBR) made from a 680 L insulated rectangular polyethylene tank with 0.45 m^3 of Kaldness® media. Marine salts and treated tap water were used. Fingerlings were acclimatized, sampled and observed for pathogenic organisms, and fed *ad libitum* with a larval ration. They were transitioned to a 2-mm Ziegler marine feed at 5% BWD and transferred to a MRAS of 62.2 m^3 with five 7,700 L circular polyethylene tanks and one 31,000 L fibreglass tank. Water treatment consisted of a 450W ultraviolet sterilizer, drum filter with 60µ screen, a 265 LPM foam fractionator and three MBBR of 2,700 L each filled with 40% Kaldness®. Water quality parameters of temperature, salinity, pH, conductivity and ammonia-nitrogen were measured daily, while nitrite-nitrogen, nitrate-nitrogen and turbidity were measured weekly. The study established that cobia could be grown utilizing sea salts but care should be taken with source of seed stock and pre-treatment of source water. Good fish health management practices, trained technical staff, and good system design are also critical elements for success.

KEY WORDS: Cobia, system design, marine recirculating aquaculture system

INTRODUCTION

Cobia are large, migratory, coastal pelagic fish and the only member of the family Rachycentridae (Holt et al. 2007). They are distributed worldwide in tropical and subtropical seas, except for the eastern Pacific (Briggs 1960, Shaffer and Nakamura 1989). They are found in a variety of habitats including mud, sand and gravel bottoms, over coral reefs, and off rocky shores (Collette 1999). They are also found in mangrove sloughs, inshore around pilings and buoys, and offshore around drifting and stationary objects, and occasionally in estuaries (Shaffer and Nakamura 1989), and are also caught for sport (Smith 1997). They feed on crabs, fishes, and squids (Collette 1999, Randal 1996). Adults reach lengths of up to two meters and weights of 60 kg, and these rapid growth rates (Chou et al. 2001), disease resistance, acclimation to tank and net pen confinement, and adaptability to commercially available aquafeeds (Schwarz et al. 2004) have all advanced its aquaculture potential.

Adult cobia, *Rachycentron canadum,* was considered one of the most promising candidates for warm-water marine fish aquaculture in the world (Liao et al. 2004) and a finfish species with emerging global potential for mariculture (Holt 2007). Benetti et al. (2010) suggested that cobia aquaculture can be viable in the Americas and the Caribbean in the next few years. Through the research programme of the University of Miami, Florida eggs, larvae, fingerlings, juveniles and brood-stock have been shipped to numerous universities, institutions and private companies, supporting research and the development of cobia aquaculture (Bennetti et al. 2010). Commercial aquaculture project locations are currently in the Bahamas, Belize, the Dominican Republic, Mexico, Philippines, Puerto Rico, United States, and Viet Nam. There is also successful cage fish farming of cobia in Panama (Benetti et al. 2010).

The global trend of aquaculture development gaining importance in total fish supply has remained uninterrupted and will continue to rise with an annual growth of 1.3% to an estimated 181 million tonnes in 2022 (FAO 2014). Aquaculture is expected to provide most of the increase in fish production with the outlook for venturing into and remaining in aquaculture highly favourable. Fisheries and aquaculture contribute to food supplies, incomes and healthy diets for millions of people all over the world and are particularly important in poverty alleviation, food security, and nutritional well-being of many coastal and rural communities in developing countries (FAO 2010). Increases in the worldwide consumption of fish and increased regulatory pressure have forced the aquaculture industry to develop more innovative approaches in production of aquatic organisms and management of the quality of effluents. One of the growing fields in aquaculture both in terms of research and commercial activities is recirculating aquaculture systems (RAS). Some challenges include limitations in quality and quantity of water, availability and cost of land, and limitations on water discharge and environmental impacts. These RAS systems reduce water usage, reuse water for continuous production, as well as recycle water for other production systems in agriculture.

Trinidad and Tobago is predominantly a natural gas and oil-based economy with petrochemical industries providing over 44% of the gross domestic product (Longmore et al. 2014). Aquaculture is still an emerging industry although there have been some successes in small-scaled fish farms and in ornamental fish breeding, farming and exports. However, increased demand for fish and fish products, coupled with declining and potential declining stocks as well as the need for diversification in the local economy have seen a renewed interest in aquaculture as a source for fish and as a potential business. The Seafood Industry Development Company (SIDC) is a state project implementation agency established by the Government of Trinidad and Tobago to partner with stakeholders to ensure that the seafood industry attains viability on a sustainable basis in the shortest possible time. The establishment of a pilot marine fish farm is one of the SIDC's initiatives to promote and develop the local aquaculture industry. This pilot farm would culture marine species in intensive re-circulating production systems to produce high priced, high value fish and products. The broader vision is to utilize the experiences gained for the development of the local aquaculture industry with the involvement of farmers, private investors, and entrepreneurs, as well as assisting in the diversification of the economy for sustainable food production.

The purpose of this study was to determine the technical feasibility and associated challenges of growing cobia in a land-based marine recirculating aquaculture system (RAS) and to create the opportunity to develop a commercial aquaculture industry based on experiences and competences acquired.

METHODOLOGY

Quarantine Recirculating Aquaculture System

Approximately 800 cobia fingerlings, 79 days post-hatch (DPH) and 2.8 ± 0.9 g were transported from a hatchery in Florida, USA to the Marine Aquaculture Facility at the SIDC in Charlieville, Trinidad. The fish were acclimated and randomly distributed into the three quarantine tanks in a marine recirculating system (QRAS). The QRAS comprised of three 2,800 L linear low-density polyethylene (LLDPE) tanks supplied with air from a 1.5 HP blower (Sweetwater®, Apopka, FL, USA) with three 7.5 cm air diffusors in each tank. The QRAS filtration system comprised of a 680 L polyethylene tank as a mechanical filter filled with approximately 0.34 m^3 of K1 media (Kaldness Inc. Providence, RI, USA) and seeded with nitrifying bacteria (Proline, Richmond, BC, Canada). A protein skimmer (All Seas Marine Model G-5, Terrance, CA, USA) was used for particulate removal and two 110 W ultraviolet sterilizers (Emperor Aquatics, Pottstowm, PA, USA) were used in series for the reduction of bacterial load. A 680 L insulated rectangular polyethylene tank with 0.45 m^3 of K1 media was used as a moving bed bioreactor (MBBR) for ammonia and nitrite removal (Figure 1). Water was recirculated with the use of a 0.5 HP high efficiency marine pump (Sweetwater®, Apopka, FL, USA) through 5 cm dia. schedule 40 PVC pipes and valves. Drain-lines were sewer-grade 10 cm dia. PVC pipes.

Municipal water supply was treated using 5µ mechanical cartridge filters and activated carbon for the removal fine particulates and possible pollutants, respectively. Appropriate amounts of sodium thiosulphate were used to remove chlorine and chloramines based on preliminary samples. Also, the addition of a water conditioner (ClorAm-X® Campbell, CA, USA) was necessary for the removal of ammonia before mixing with commercial marine salts. The total volume of seawater in the system was 10,440 L.

Fish were exposed to a 12:12 light:dark cycle with marine blue LED lights controlled by digital timers position 2 m above the culture tanks.

Fingerlings were hand-fed *ad libitum* daily for the first 23 days with Zeigler AP100 commercial larval ration containing a minimum of 50.0% crude protein and 12% crude fat. This was also supplemented with recently-hatched brine shrimp nauplii, *Artemia salina*. They were then fed a commercial pellet containing a minimum of 50% crude protein and 15% crude fat (Zeigler Finfish Starter with VpakTM) at a reduced rate from 10.0 to 5.0 % average body weight (ABW)/day throughout the quarantine period. Growth was evaluated as specific growth rate (SGR) expressed in percentage per day (%/d), described by the formula:

$$SGR = [(\ln(W_f) - \ln(W_i)] \times \frac{100}{t}$$

where W_f, final weight (g), W_i is initial weight(g) and t, time between measurements (d). Feed conversion ratio (FCR) was calculated for each sample period to express the relationship between feeding rate and SGR (Orellana *et al.* 2013).

Fish were also sampled and necropsies done for observation of pathogenic organisms and general fish health management.

Marine Recirculating Aquaculture System

At the end of the quarantine period, the cobia fingerlings were transferred to a marine recirculating aquaculture system (MRAS1) after receiving a prophylactic treatment of 37 - 40% formaldehyde at 150 mg/L for approximately 15 minutes. The MRAS1 comprised of five LLDPE tanks of 7,700 L each with one used as a sump and one 31,000 L fibreglass tank supplied with air from a 5 HP blower (Sweetwater® Apopka, FL, USA) via rubber-polyethylene grid diffusers. The filtration system consisted of an

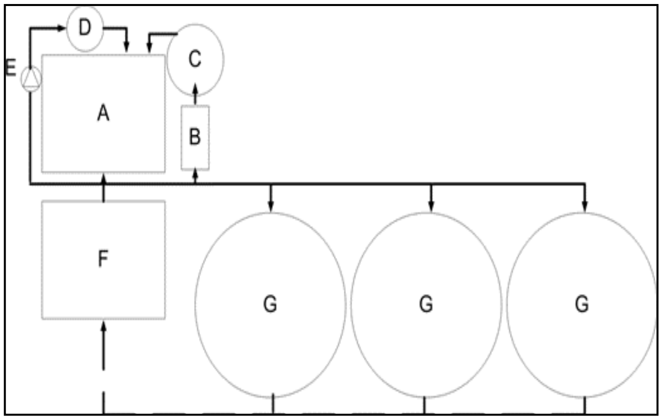

Figure 1. Process flow of quarantine recirculating aquaculture system used for pilot study at the SIDC Facility in Trinidad and Tobago. Key components are (A) Sump, (B) UV sterilizer, (C) moving bed bioreactor, (D) foam fractionator (E) pump (F) mechanical filter, (G) tanks.

automatic backwash 1,500 LPM drum filter with a 60 µ screen panel (Faivre Drum Filter 2-80 Series, Baume-les-Dames, France) and a 265 LPM foam fractionator (RK2, San Diego, CA, USA) for the removal of large and small particles, respectively. A high-intensity 450 watts, 969 LPM ultraviolet sterilizer (Emperor Aquatics, Pottstown, PA, USA) was installed for reduction of bacterial load. Three MBBR of 2,700 L each were used for ammonia and nitrite removal using K1 media filled to approximately 4.01m^3 each and seeded with nitrifying bacteria. Each MBBR was supplied with four 23 cm disc diffusers (FlexAir® Paris Rd, Columbia) with 1.27 cm PVC valves to control air flow and movement of the beads. One MBBR was used primarily for the removal of carbon dioxide via heavy aeration and an extractor fan while the other two were in series for nitrification. Two marine pumps, a 0.5 HP high efficiency (Sweetwater®, Apopka, FL, USA) and a 2 HP Wave II high speed (W. Lim Corporation, Mira Loma, CA, USA) were used to recirculate the water volume of 77,600 L in the system.

Municipal water supply was treated using 5µ mechanical cartridge filters and activated carbon for the removal of fine particulate and possible pollutants. Appropriate amounts of sodium thiosulphate were used to remove chlorine and chloramines based on preliminary samples. Also, the addition of a water conditioner (ClorAm-X® Campbell, CA, USA) was necessary for the removal of ammonia before mixing with commercial marine salts. To reduce the cost a combination of marine rock salt together with the commercial marine-mix salts were mixed to achieve desired salinity and conductivity. Trace elements such as Zinc, Iron, Manganese and Cobalt were added. (Figure. 2).

Water Quality

Salinity, temperature, and conductivity were measured with a YSI meter (YSI® 30 Yellow Springs, OH, USA) and pH was measured with a Hanna® pH Pen (Woonsocket, RI, USA). Dissolved oxygen was maintained above 6.0 mg/L. Total ammonia nitrogen TAN (Salicylate method), nitrite-nitrogen NO_2-N (Diazotization method), nitrate-nitrogen NO_3-N (Cadmium reduction) and turbidity (Absorptiometric method) were measured using a SMART3 Colorimeter (LaMotte® Chestertown, Maryland, USA). Unionized ammonia (UIA) was calculated based on TAN, pH, and temperature. Salinity, conductivity, temperature, and pH were measured twice daily. Total ammonia nitrogen, nitrite-nitrogen, nitrate-nitrogen, and turbidity were measured at least once per week or when required. Fish were exposed to a 12:12 light:dark cycle with marine blue LED lights controlled by digital timers positioned 2 m above the culture tanks.

Feeding

Fish were hand-fed four times daily between the hours of 08.00 to 16:00. The ration was divided equally among the four feeding based on a reduced rate from 5.0 to 3.0 % average body weight (ABW)/day using a commercial pellet containing a minimum of 50% crude protein and 15% crude fat (Zeigler Finfish Starter with VpakTM, Pennsylvania, USA).

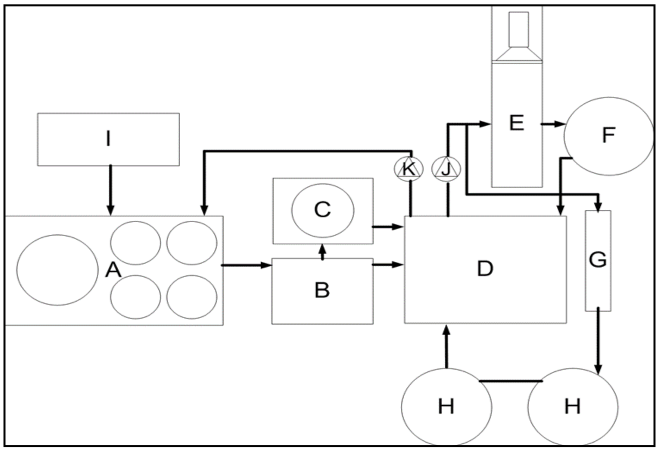

Figure 2. Process flow of quarantine recirculating aquaculture system used for pilot study at the SIDC Facility in Trinidad and Tobago. Key components are (A) Sump, (B) UV sterilizer, (C) moving bed bioreactor, (D) foam fractionator (E) pump (F) mechanical filter, (G) tanks.

Sampling

Fish were sampled and the length and weight were measured using a stainless steel measuring tape and a bench scale (3 kg ± 0.02) (Ohaus, Parsippany, NJ, USA) respectively. They were also observed for any signs of abnormality or pathogenic organisms. This was done via scrapes of the external body surface, fin and tail clips, gill examination and necropsies of dead or moribund specimens.

RESULTS

Total Ammonia-Nitrogen (TAN) concentrations in the QRAS averaged 0.96 ± 1.12 mg/L (max. 4.50 mg/L and min. 0.04 mg/L) and unionized ammonia (UIA) concentration averaged 0.08 ± 0.19 mg/L (max. 1.00 mg/L and min 0 mg/L) during the first 98 days of the cobia trial (Figure 3). Elevated TAN concentrations of above 1.0 mg/L were observed during the first 27 days of the trial. Subsequently, TAN concentrations remained relatively low. The average concentration of nitrite-nitrogen was 19.64 ± 8.91 mg/L (max 46.50 mg/L and min 0.20 mg/L) (Figure 3.).

After the acclimation period, salinity levels of the QRAS averaged 30.32 ± 0.70 g/L (max 31.63 g/L and min 28.73 g/L) while daily water temperature averaged at 25.07 ± 1.11°C (max 29.19°C and min 22.15°C) in the QRAS (Figure 4). The average daily pH values were 7.69 ± 0.33 (max 8.27 and min 7.08). The pH levels were initially elevated up to day 21, after which they gradually dropped for the remaining time the cobia were in quarantine (Figure 4).

Figure 5 highlights the TAN concentration in the MRAS which averaged 0.76 ± 0.68 mg/L (max 1.78 mg/L and min 0.01 mg/L). Initially, there was a daily sharp increase in TAN, peaking on day 12 at 1.78 mg/L after which the concentration dropped and remained stable at approximately 0.05 mg/L. The UIA of the MRAS averaged 0.03 ± 0.03 mg/L (max 0.07 mg/L and min 0 mg/L) where the concentrations were elevated for the first 17 days in MRAS with subsequent concentrations being stable at 0 mg/L (Figure 5). Nitrite-nitrogen concentrations in the MRAS (Figure 5) averaged at 4.42 ± 4.06 mg/L (max 10.05 mg/L and min 0.02 mg/L). Generally, nitrite-nitrogen concentration showed a sharp increase from 0.02 mg/L on day 7 and peaking on the 28 day at 10.05 mg/L.

Salinity was 30.98 ± 1.11 g/L (max 32.20 g/L and min 28.90 g/L) over the quarantine period. The cobia entered the MRAS1 at the salinity level of 28.9 g/L and salinity levels were stable for the first 28 days in MRAS. Conductivity levels averaged of 48.53 ± 2.01µS/cm (max 50.88 µS/cm and 44.46 µS/cm). The values for pH in the MRAS averaged 7.87 ± 0.05 (max 7.97 and min 7.76). Water temperature in the MRAS averaged 26.4 ± 0.5 °C (max 27.3 °C and min 25.4 °C). Daily salinity, conductivity, pH and temperature parameters are shown in Figure 6.

Figure 7 highlights the growth of cobia during the study period. The average weight increased from the 2.8 ± 0.9 g when the cobia were stocked into Quarantine to 22.97 ± 5.58 g when they were stocked in the MRAS to 26.85 ± 4.52 g at the end of the 125 day study period.

Figure 8 highlights the pH of the domestic water supply averaged 6.48 ± 0.36 (max 7.43 and min 5.85) over a 30 day period.

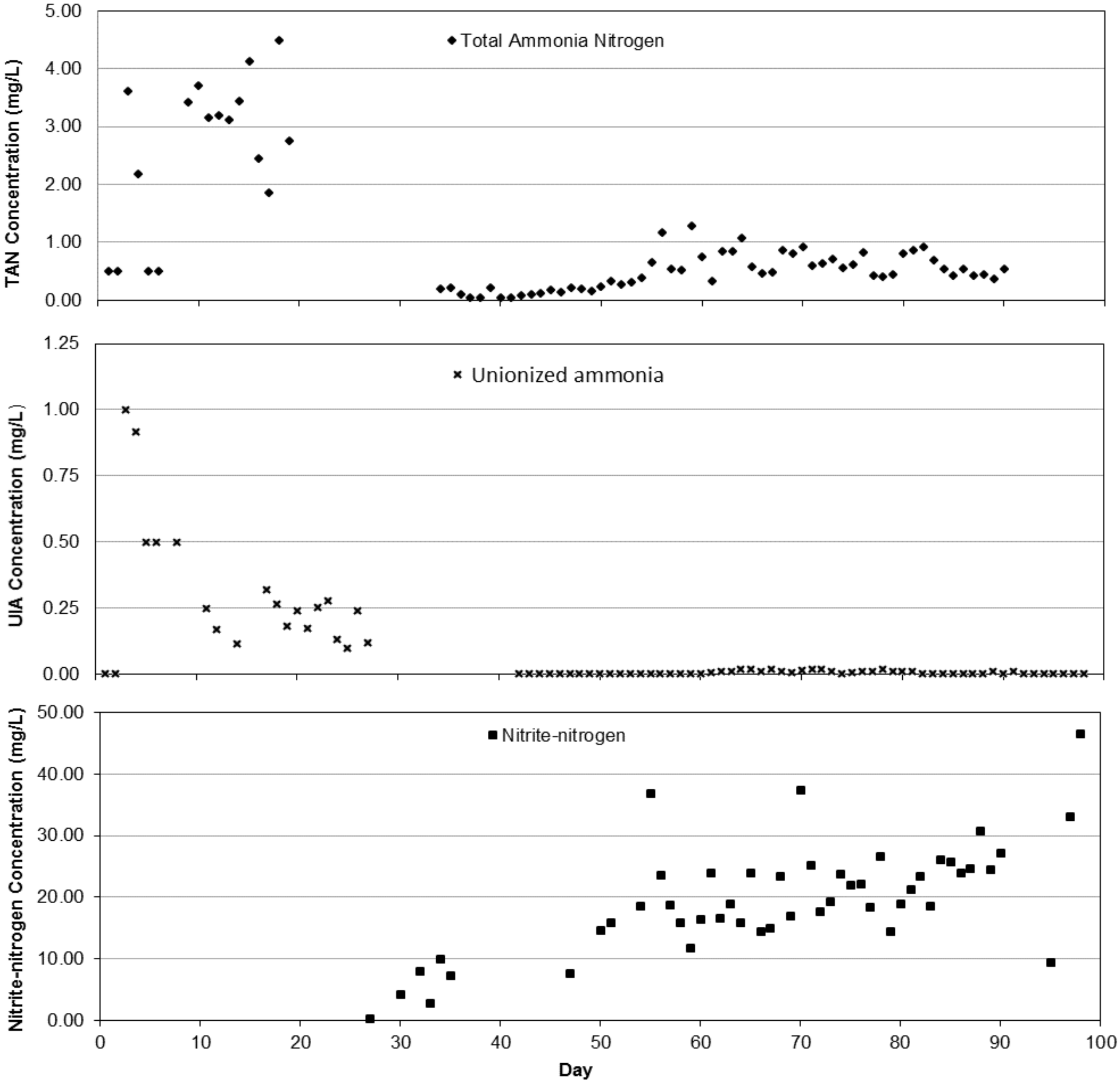

Figure 3. Total ammonia nitrogen, unionized ammonia and nitrite-nitrogen during the cobia pilot study at the SIDC Quarantine

DISCUSSION

Water quality management was one of the factors that played a crucial role in the culture of cobia in this study. Water quality management in a new marine recirculating system proved to be a challenge, especially in the quarantine after initial stocking of fish with *ad libitum* feeding. Increases in TAN and nitrite followed classical patterns of nitrogenous concentrations in new systems (Ebeling 2006) despite seeding of biofilters with commercial nitrifying bacteria. In the initial stocking phase, when the cobia were first in quarantine, there were fluctuations in the total ammonia-nitrogen and a peak of 4.20 mg/L. The TAN was controlled by frequent water changes until additional active bio-media was added to the system to control and manage the high levels of ammonia. The UIA is also affected by temperature and pH and elevated values were recorded in the first 28 days of the trial because of pH levels above 8.0. From day 41 until being transferred out of the QRAS1, the UIA remained stable below 0.02 mg/L. Nitrite-nitrogen levels increased to a high of 46.40mg/L.

Salinity was increased incrementally from the shipping salinity of 14.60 g/L to 30.00g/L and was maintained at that salinity until the end of the quarantine period. Water added to the system was primarily for water exchanges when there were ammonia spikes. Pipe-born water is a cheap and convenient source for mixing sea-water when marine salt is added but may cause problems if the source is not reliable and will present serious biosecurity concerns with its potential to accommodate harmful pathogens. In addition, inconsistent quality, heavy sediment loads and the presence of contaminants including ammonia, phosphates and nitrates also provided challenges and increased costs for water treatment. This is most evident with the fluctuation of pH highlighted in Figure 8. Efforts to lower costs by incorporating lower-cost salt combinations may have also compounded problems. Growing cobia in marine recirculating systems is technically viable but includes some challenges. Benetti et al. (2008) suggest that researching the potential for culturing cobia is necessary because despite their relative hardiness, they are suscepti-

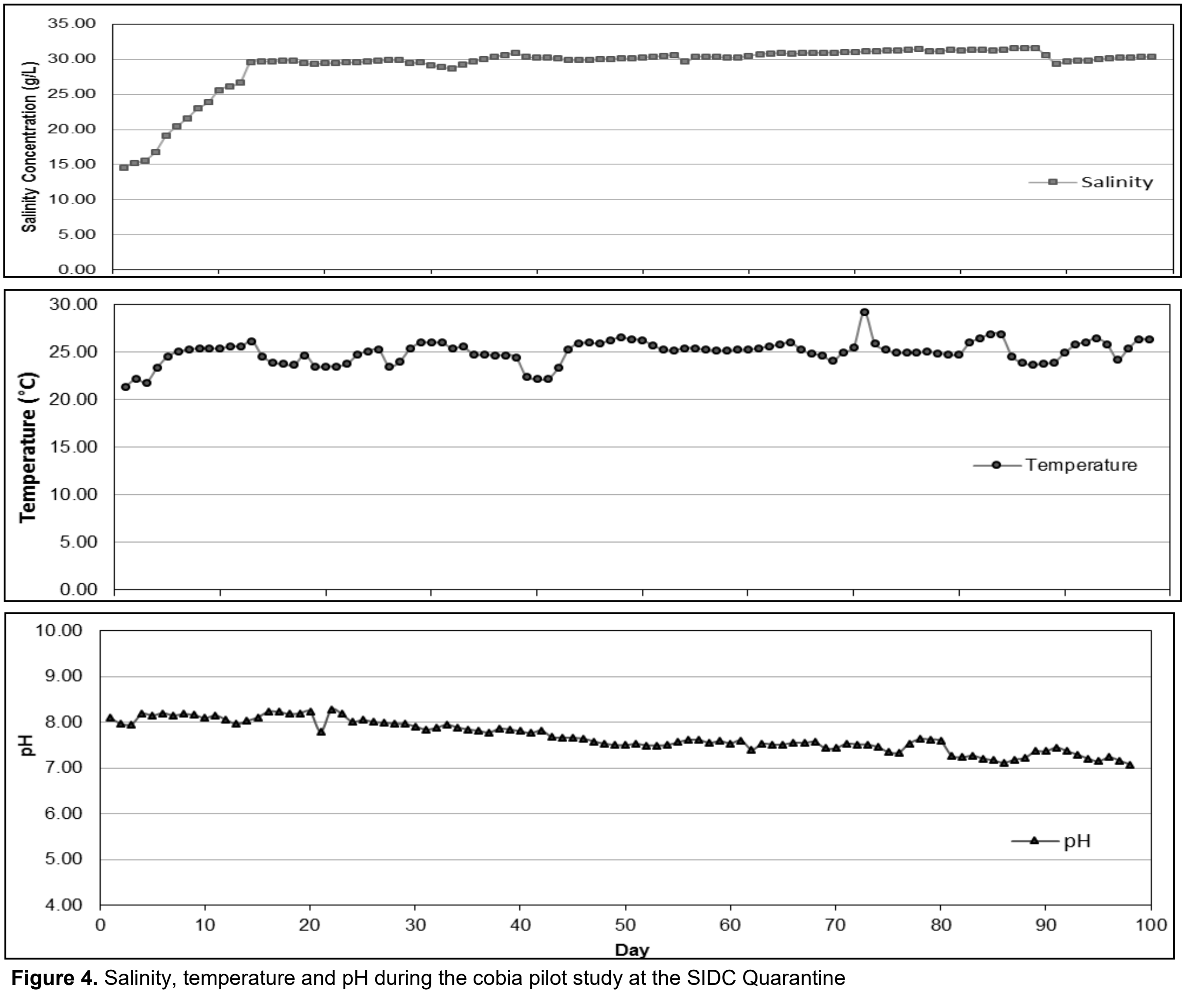

Figure 4. Salinity, temperature and pH during the cobia pilot study at the SIDC Quarantine

ble to a wide range of pathogens, including viruses such as *Lymphocystis*, bacteria such as *Vibrio spp.*, and various types of parasites. The larvae are also particularly vulnerable to outbreaks of bacterial enteritis and bacterial gill disease. In this pilot study, cobia fry showed signs of gill hyperplasia, a protozoan parasite *Amyloodinium sp.* as well as a systemic bacterial infection which caused some mortalities and was successfully treated. However, this prolonged the quarantine period as great care was taken to successfully treat the animals before they were transferred for growout. These various stressors may have affected growth.

This emphasizes the importance of the source stock of seed introduced into marine production systems and underlines the importance of proper quarantine and fish health management facilities. Proper support from qualified professionals in fish health management and aquatic medicine is also important for success. Staff with limited technical skill and knowledge may also affect operations, therefore hands-on training and in-house seminars are important to develop appropriate levels of skill, experience and competences in the different areas of expertise required to deal with challenges of managing marine recirculating systems.

The pilot study demonstrated that marine recirculating systems offer viable means of production for marine fish but care must be taken in the sourcing of seed stock, proper quarantine and fish health management protocols, trained staff and pre-treatment of all water and salts used in the manufacture of the seawater for growing the animals. With proper planning and scheduling of activities including critical paths for equipment procurement, biofilter seeding and stabilization, with healthy animal introduction to growout facilities, successful production can be achieved.

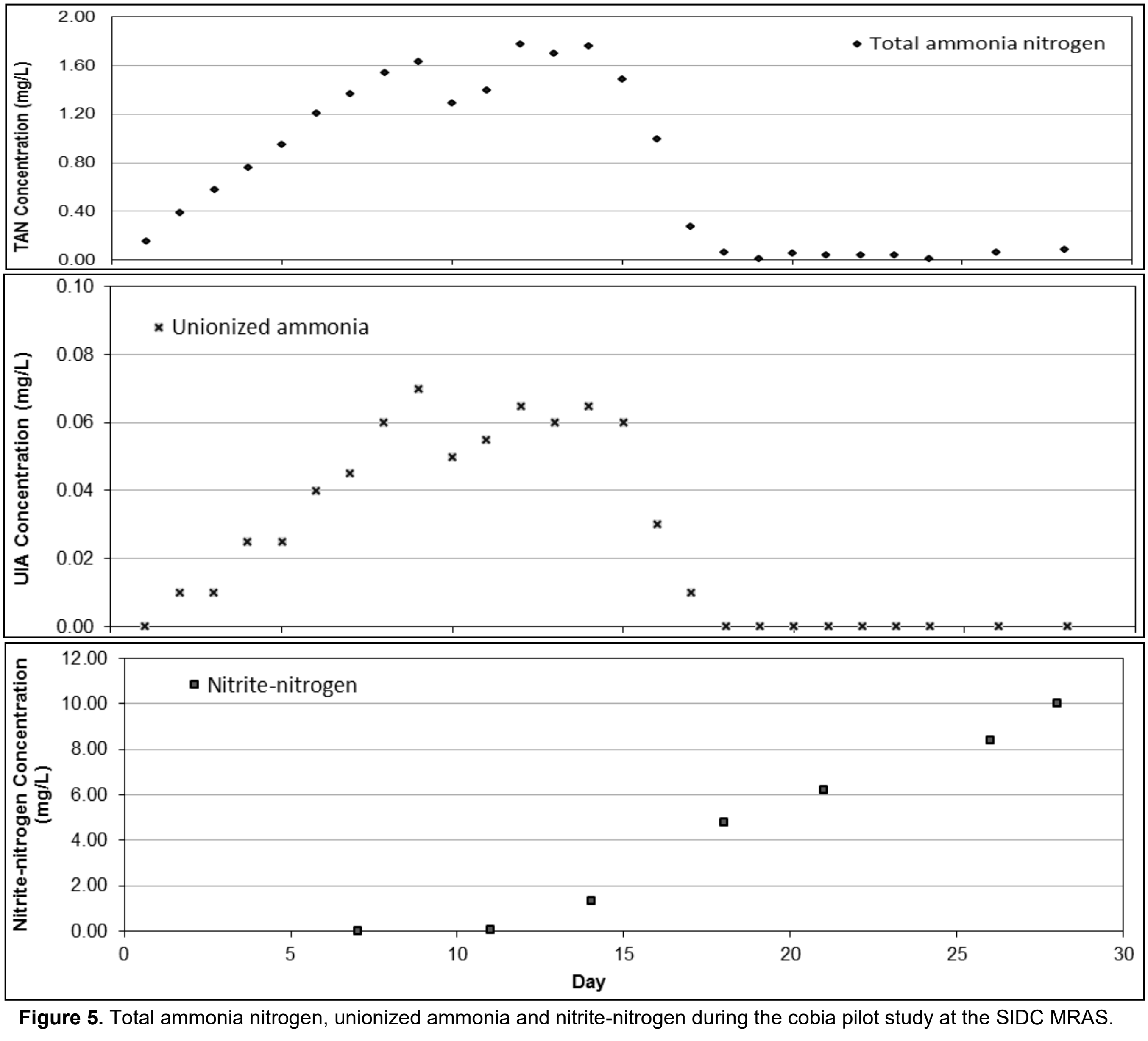

Figure 5. Total ammonia nitrogen, unionized ammonia and nitrite-nitrogen during the cobia pilot study at the SIDC MRAS.

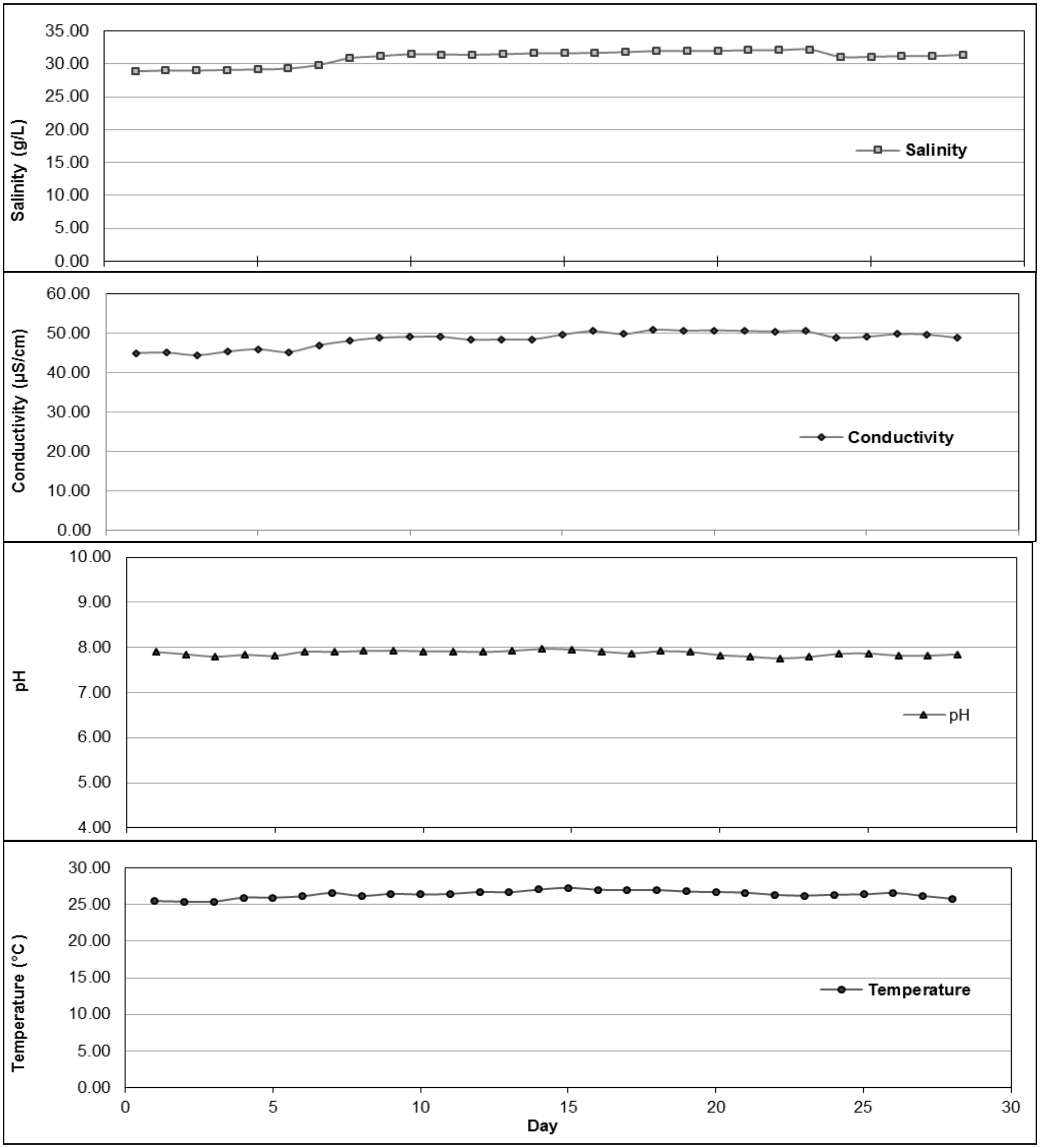

Figure 6. Salinity, conductivity, pH and temperature during the cobia pilot study at the SIDC MRAS

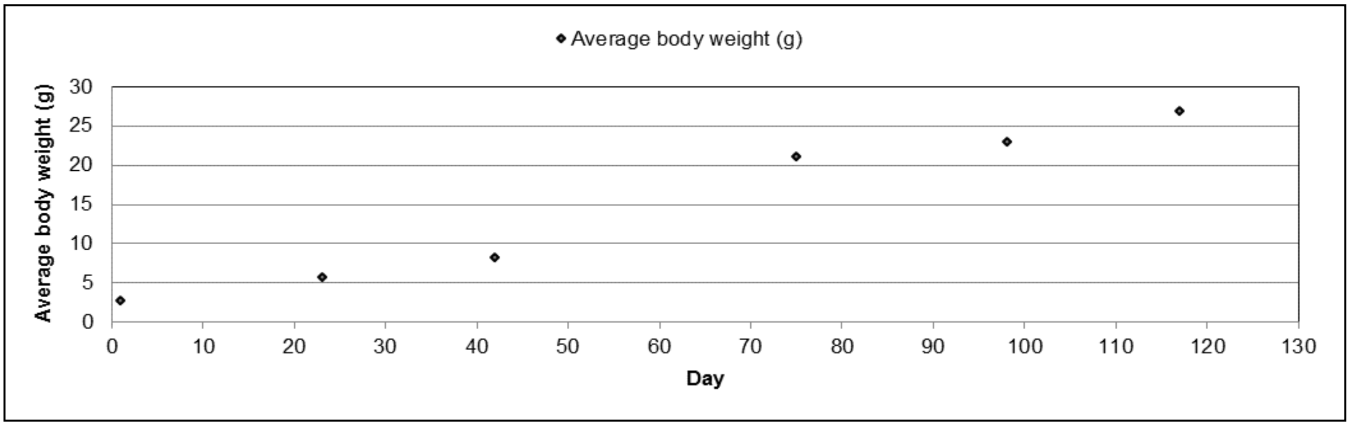

Figure 7. Average body weight of cobia

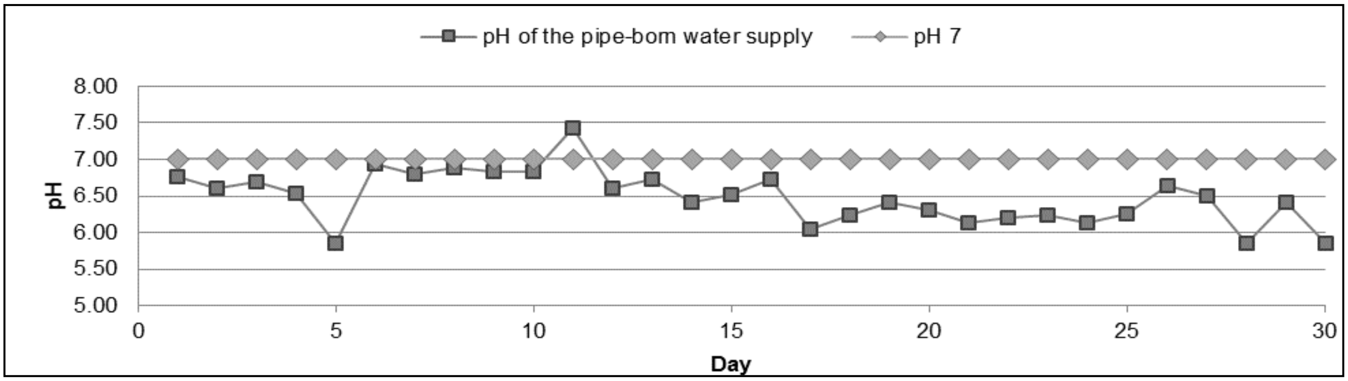

Figure 8. Fluctuating pH values of the pipe–born water supply

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