Effect of partial feed deprivation on plasma total protein and glucose as stress indicators in African catfish *Clarias gariepinus* (Burchell 1822)

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ABSTRACT

A 28 days study was conducted to determine the relationship between partial feed deprivation and stress in African catfish Clarias gariepinus, using plasma total protein and glucose as stress indicator. A total of 120 catfish with an initial weight of 200 ±10 g were randomly stocked in triplicate at 20 fish per group and fed twice daily with a commercial diet at 9:00 am and 4:00 pm. The control groups (labelled B) were fed with 4% of their body weight while the experimental group (labelled A) were fed with 1% of their body weight. Blood samples were obtained from 30 fishes (15 from each group at 5 per tank) at intervals of 7 days for the entire experimental period and plasma total protein and glucose were measured. Although plasma total protein and glucose were not affected by feeding strategy at day 7, but are slightly lower compared to the control group. Furthermore, a significant decreased was observed at day 14, 21 and 28 in the experimental fish, which were most evident of stress in those fishes subjected to 28 days feed deprivation, unlike those of control groups that revealed more of normal plasma total protein and glucose levels. The study has observed that varying degree of starvation in catfish could have accounted for the stress in African catfish, Clarias gariepinus.

Keywords: Partial feed deprivation, Plasma biochemistry, Total protein, Glucose, African catfish,

INTRODUCTION

Feed deprivation is stated were by feed is withdrawn in a designed experimental way (Costas et al., 2011). Fish can survive a long time without feed, and fasting is a natural part of many lifecycles (Navarro & Gutiérrez, 1995). It is known that many fish alternate feeding and feed deprivation periods during the annual cycle in nature as a consequence of reproductive processes or seasonal variations in temperature or feed availability (Gaylord & Gatlin III, 2000). To survive such feed restrictions, fish mobilize their energy reserves which impose metabolic adjustments that are species dependent (Navarro et al., 1997). Intraspecific variability of the responses of fish to feed deprivation is huge, and several biotic (age, size, feeding and health status prior to feeding deprivation) and abiotic (e.g., season, temperature, salinity) factors are of profound importance in setting the stage on which the reorganization of metabolism can take place (Iii, 2001; Costas et al., 2011). Fishes, like their mammalian counterparts, are affected by lack of adequate feed supply, and this will surely manifest in them as stress (Martinez et al., 2009; Polakof et al., 2012).

Stress in fish is an unavoidable consequence of normal practices in aquaculture. In extreme cases, a variety of stressors, including capture and handling, crowding, and nutritional state or feeding strategy, which may have negative effects on reproduction, body shape, growth rate and flesh quality, and susceptibility to disease (Waagbo et al., 1994;Mountfort et al., 2002; Li, Li, Zhang, & Tao, 2013). The primary physiological response to such perturbations is endocrine in origin where rapid, transient adrenergic flushes give way to elevated and sustained corticosteroid levels when stress persists as a chronic condition (Cui et al., 2010; Martinez-Porchas et al., 2009). While it remains a complex and specialized exercise to monitor these primary hormonal events, their secondary effects in altering the homeostatic balance of metabolism may be perceived through monitoring a range of blood chemistry and haematological factors.

Recent development in the study of stress responses in fish has shown that blood glucose, lactate, and plasma protein values correlate with a wide range of stressors (Sinha et al., 2011; Pottinger, 1998). The development of methods for monitoring metabolic indicators of stress in fish, therefore, has obvious potential for improving husbandry protocols and post-harvest product quality. This approach has the practical advantage that blood sampling is nondestructive and relatively simple to carry out. Nevertheless, the feasibility of detecting and monitoring stressed states in fish under field conditions has been investigated by Morgan et al. (1997) both concluded that blood glucose analysis with portable instrumentation provided reliable measures of stress and had strong potential for use in aquaculture and field monitoring facilities. The aim of the present study is to establish the role of partial feed deprivation (if any) in causing stress to African catfish *Clarias gariepinus* through the measuring of plasma total protein and glucose as stress indicators.

MATERIALS AND METHODS

Experimental animals, acclimation and experimental design

This experiment was carried out in the Department of Biology, Faculty of Science Abubakar Tafawa Balewa University Bauchi Nigeria. Active, live *Clarias gariepinus* (120) of about 200-210 grams body weight were used for the investigation. They were obtained from National Institute for Freshwater Fisheries Research (NIFFR) New-Bussa, Nigeria. Fish were acclimatized for 14 days before onset of the experiment. They were randomly placed into triplicated groups A (feed deprived) and B (control) of 20 each. The control group was fed twice daily, at 9:00 am and 5:00 pm, with 4% of their body weight, while the experimental group was fed once daily, at 5:00 pm, with 1/4 the quantity is given to the control groups of 6mm extruded floating commercial pellets (Coppens 42% Crude Protein) as recommended by Craig and Helfrich, (2002). This quantity and time of feeding were maintained throughout the experimental period, as irregular feeding will retard the growth rate of fish (Bolorunduro 2013).

Water quality parameters including water temperature, dissolved oxygen, pH and total ammonia were measured on each non-sampling days, and approximately 50% of the water was exchanged according to Kamarudin et al. (2011). Throughout the experiment, water temperature ranged 28.2-29.6 °C, dissolved oxygen 7.5-7.8 mg L^{-1}, pH 7.1-7.5, and ammonia-nitrogen of 0.01-1.14 mg L^{-1}.

Blood Analysis

At the end of each 7 days feeding period, five fish in each tank were randomly sampled and anaesthetised with 50 mg L^{-1} tricaine methane sulfonate (MS 222). Each fish was individually weighed and the blood was sampled by puncturing the caudal vein with a heparinised 23G syringe. The blood was centrifuged and stored in a freezer (-80 °C) for analysis according to (Sulaiman et al., 2017)

Statistical Analysis

The values obtained were expressed as mean ± SEM and tabulated. These were subjected to a Two-tailed unpaired student t-Test to compare the two feeding methods used after prior confirmation of data homogeneity using Statistical Analysis System 9.4 for Windows (SAS Inc., USA). Differences among

dietary treatments were identified using least significant difference (LSD) at $P < 0.05$.

RESULTS

Blood plasma parameters of *Clarias gariepinus* were significantly affected by the feeding strategy. Although plasma total protein and glucose were not affected by feeding strategy at day 7, but are slightly lower compared to the control group. However, a significantly decreased was observed at day 14, 21 and 28 in the experimental fish, Total protein and glucose levels in the feed deprived *Clarias gariepinus* group significantly decreased with increase in days from 14 to 28 days. (Fig. 1-6).

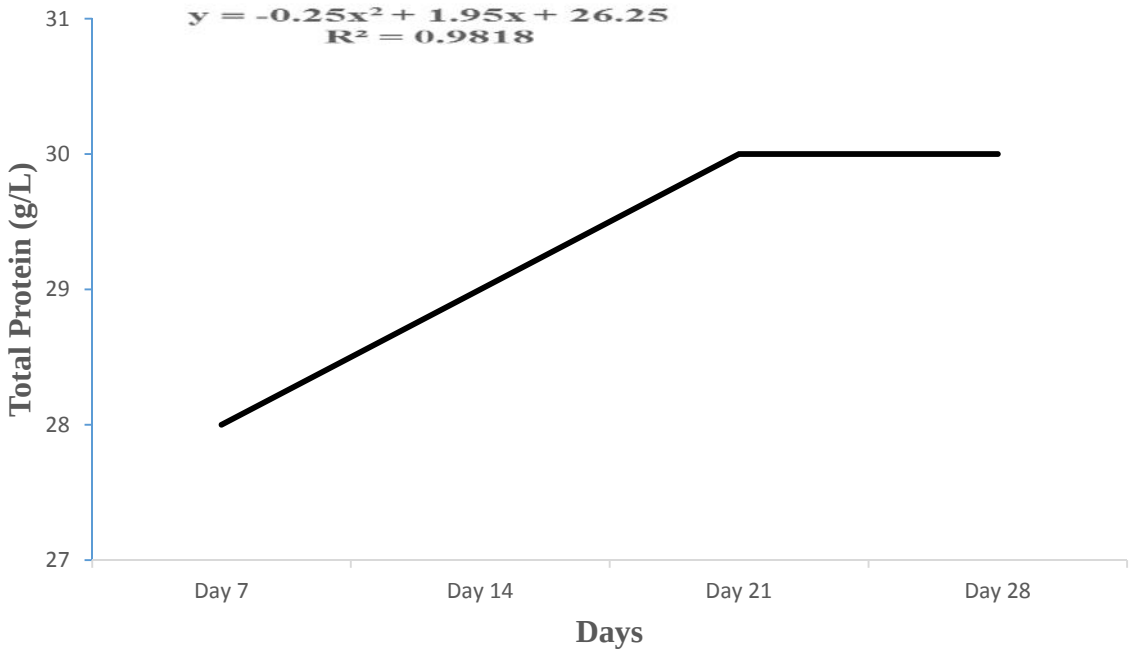

Figure 1: Relationship between control *C. gariepinus* and plasma total protein with days.

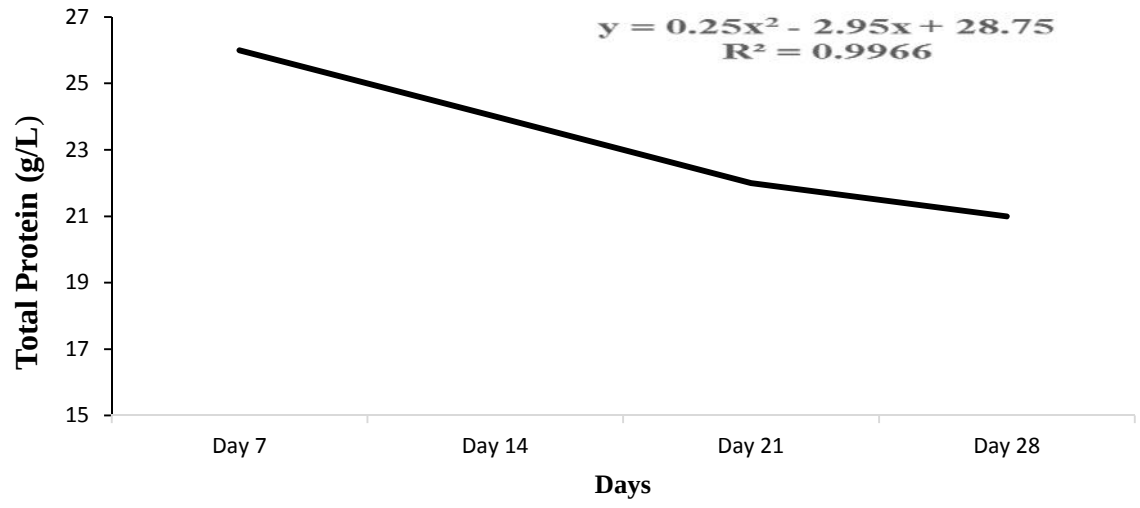

Figure 2: Relationship between feed deprived *C. gariepinus* and plasma total protein with days.

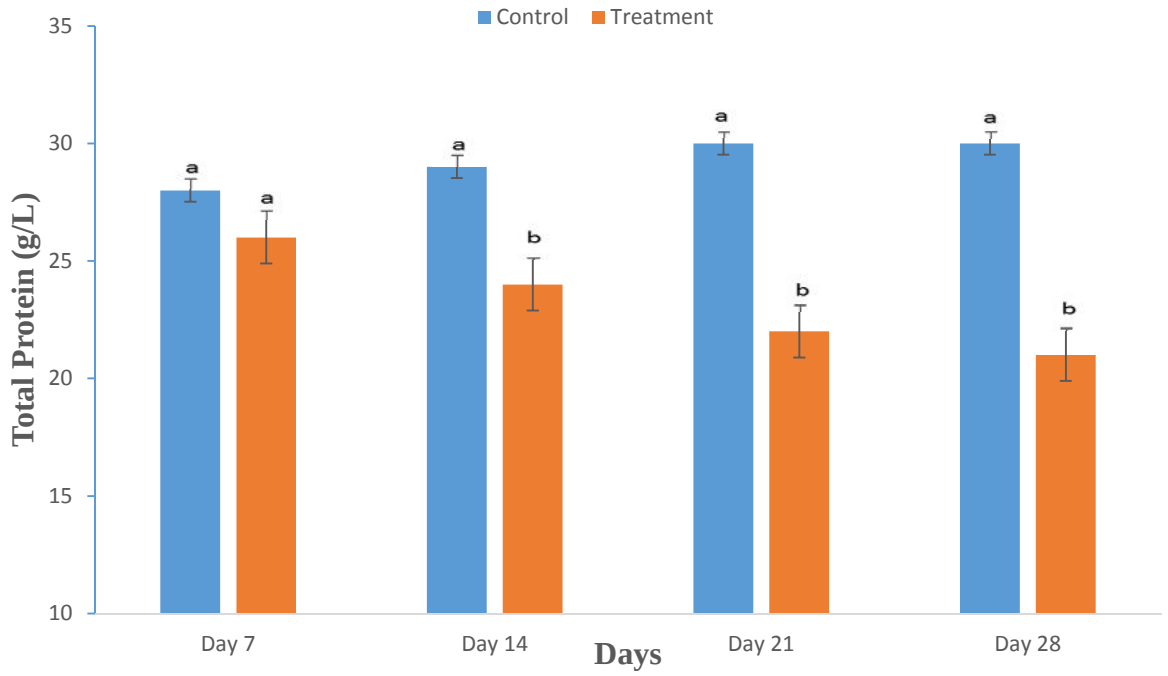

Figure 3: Relationship between controls, feed deprived group *C. gariepinus* and plasma total protein with days. Different letters in each treatment indicate significant differences ($P < 0.05$)

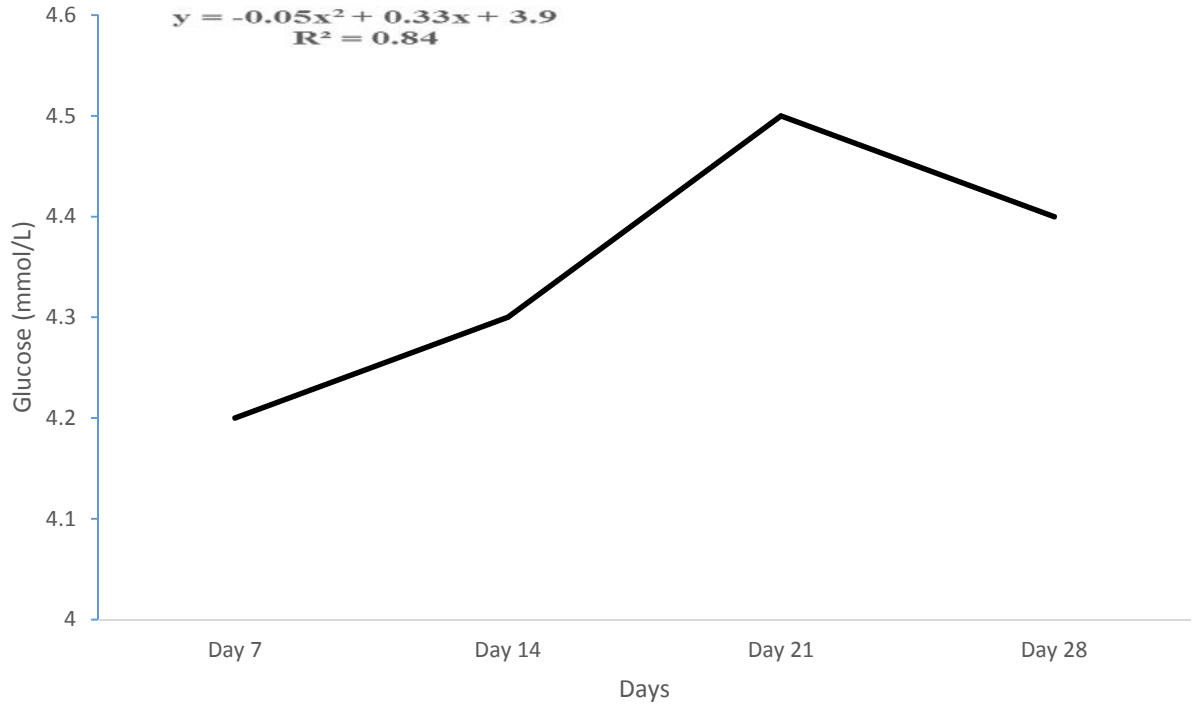

Figure 4: Relationship between control *C. gariepinus* and plasma glucose with days.

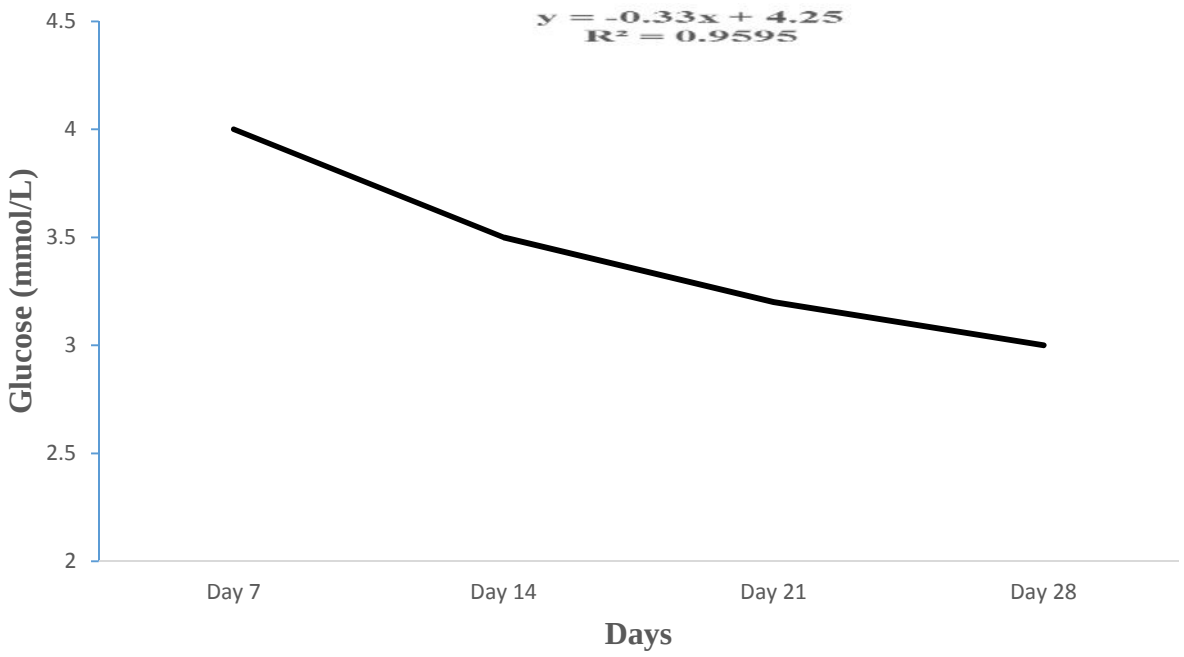

Figure 5: Relationship between feed deprived *C. gariepinus* and plasma glucose with days.

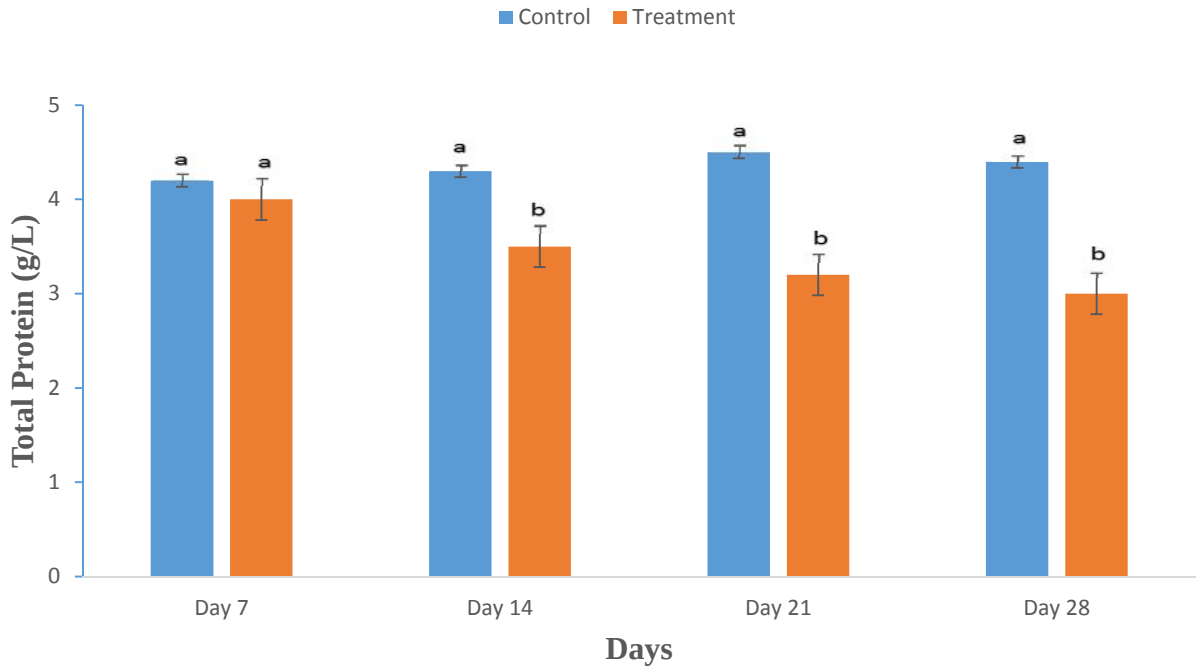

Figure 6: Relationship between controls, feed deprived group *C. gariepinus* and plasma glucose levels with days. Different letters in each treatment indicate significant differences ($P < 0.05$)

DISCUSSIONS

In the present study, feed deprivation had significantly affected plasma total protein and glucose levels. Even though the glucose level was within the clinical range but still lower than the control group. Total protein and glucose decreased with time and showed good negative linear correlation with feed deprivation. Total protein concentrations decreased in the plasma of feed deprived fishes in each of the three replicate compared to the control which is in agreement with previous studies reported (Hemre, Mommsen, & Krogdahl, 2002; Navarro & Gutiérrez, 1995; Peres & Oliva-Teles, 2002), meanwhile Navarro & Gutiérrez (1995) reported higher plasma protein within ration size. Nevertheless, plasma total protein concentrations were lower in fasted fishes than in fed fishes after 14–16 days (Chen et al., 2004). Similarly declined in total plasma protein was observed in rainbow trout and Chinook salmon (Congleton and Wagner 2006).

The level of plasma glucose at any time is a function of many factors such as diet, age, time since feeding, and season, and therefore is a more equivocal index of stress than cortisol (Polakof et al., 2012; Rotllant & Tort, 1997). However, quantification of plasma glucose can provide valuable complementary information regarding the severity and duration of the stress response, the time required for recovery from the stimulus, and provides another point of reference with previous work on the same and related species (Chen et al., 2003).

Reduction in plasma glucose levels induced by feed deprivation has been reported in some fish species (Shimeno et al., 1997; Gillis & Ballantyne, 1996). Nevertheless, glucose levels are usually maintained in feed-deprived fish either by (a) reducing rate the of glucose utilization; (b) increasing gluconeogenic and glycogenolytic potentials; and (c) enhancing liver glucose exporting capacity (Navarro & Gutiérrez, 1995). Since glucose is an essential fuel for a number of tissues, it is particularly important that glucose levels are maintained throughout a feed deprivation period (Gillis & Ballantyne, 1996). In the present study, plasma glucose levels in 21 days feed-deprived sole remained constant, suggesting that a metabolic reorganization has occurred in order to maintain glucose levels without dietary sources. However plasma glucose levels are elevated during stress in fish primarily as a consequence of elevated blood catecholamine levels, although the involvement of cortisol in glycogenolytic/gluconeogenic processes has not been discounted (Costas et al., 2011; Sulaiman et al., 2017).

CONCLUSION

These findings indicate the potential role of total protein and glucose as indicators of stress in fish which may be used to evaluate the effects of feed deprivation on *Clarias gariepinus*. The plasma total protein and glucose levels gave a true reflection of the stress. In this study, a varied degree of starvation in *Clarias gariepinus* accounted for a proportional decreased in plasma total protein and glucose.

RECOMMENDATIONS

It is apparent from this study that further works should be done to establish on the liver and liver serum enzymes, glycogen, cholesterol and triglycerides during feed deprivation could also be investigated, along with serum levels of creatinine phosphokinase in *C. gariepinus* and other cultured fish species.

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REFERENCES

Chen, C.-Y., Wooster, G. A., Getchell, R. G., Bowser, P. R., & Timmons, M. B. (2003). Blood chemistry of healthy, nephrocalcinosis-affected and ozone-treated tilapia in a recirculation system, with application of discriminant analysis. *Aquaculture*, *218*(1), 89–102.

Chen, C. Y., Wooster, G. A., & Bowser, P. R. (2004). Comparative blood chemistry and histopathology of tilapia infected with Vibrio vulnificus or Streptococcus iniae or exposed to

carbon tetrachloride, gentamicin, or copper sulfate. *Aquaculture*, *239*(1–4), 421–443.

Congleton, J. L., and Wagner, T. (2006). Blood-chemistry indicators of nutritional status in juvenile salmonids. *Journal of Fish Biology*, *69*, 473–490.

Costas, B., Arag, O. C., Ruiz-Jarabo, I., Vargas-Chacoff, L., Arjona, F. J., Dinis, M. T. (2011). Feed deprivation in Senegalese sole (*Solea senegalensis* Kaup, 1858) juveniles: Effects on blood plasma metabolites and free amino acid levels. *Fish Physiology and Biochemistry*, *37*(3), 495–504.

Cui, X.-J., Zhou, Q.-C., Liang, H.-O., Yang, J., & Zhao, L.-M. (2010). Effects of dietary carbohydrate sources on the growth performance and hepatic carbohydrate metabolic enzyme activities of juvenile cobia (*Rachycentron canadum* Linnaeus). *Aquaculture Research*, *42*(1), 99–107.

Gaylord, T. G., & Gatlin III, D. M. (2000). Assessment of compensatory growth in channel catfish *Ictalurus punctatus* R. and associated changes in body condition indices. *Journal of the World Aquaculture Society*, *31*(3), 326–336.

Gillis, T. E., & Ballantyne, J. S. (1996). The effects of starvation on plasma free amino acid and glucose concentrations in lake sturgeon. *Journal of Fish Biology*, *49*(6), 1306–1316.

Hemre, G. I., Mommsen, T. P., & Krogdahl, A. (2002). Carbohydrates in fish nutrition: Effects on growth, glucose metabolism and hepatic enzymes. *Aquaculture Nutrition*, *8*(3), 175–194.

Iii, D. M. G. (2001). *Nutrition and Fish Health*. (C. L. C. D. Webster, Ed.), *Fish Nutrition* (1st ed.). London: The Haworth Press.

Kamarudin, M. S., Otoi, S., & Saad, C. R. (2011). Changes in growth, survival and digestive enzyme activities of Asian redtail catfish, *Mystus nemurus*, larvae fed on different diets. *African Journal of Biotechnology*, *10*(21), 4484–4493.

Li, X. L., Li, G., Zhang, S. Y., & Tao, L. (2013). Effect of recirculating aquaculture system (RAS) on growth performance, body composition and hematological indicators of allogynogenetic crucian carp (*Carassius auratus gibelio*). *Advance Journal of Food Science and Technology*, *5*(3), 348–355.

Martinez-Porchas, M., Martinez-Cordova, L. T., & Ramos-Enriquez, R. (2009). Cortisol and Glucose : Reliable indicators of fish stress ? *Journal of Aquatic Sciences*, *4*, 158–178.

Morgan, J. D., Sakamoto, T., Grau, E. G., & Iwama, G. K. (1997). Physiological and respiratory responses of the Mozambique tilapia (*Oreochromis mossambicus*) to salinity acclimation. *Comparative Biochemistry and Physiology Part A: Physiology*, *117*(3), 391–398.

Mountfort, D. O., Campbell, J., & Clements, K. D. (2002). Hindgut fermentation in three species of marine herbivorous fish. *Applied and Environmental Microbiology*, *68*(3), 1374–1380.

Navarro, I., Blasco, J., Banos, N., & Gutierrez, J. (1997). Effects of fasting and feeding on plasma amino acid levels in brown trout. *Fish Physiology & Biochemistry*, *16*(4), 303–309.

Navarro, I., & Gutiérrez, J. (1995). Fasting and starvation. *Biochemistry and Molecular Biology of Fishes*, *4*, 393–434.

Peres, H., & Oliva-Teles, A. (2002). Utilization of raw and gelatinized starch by European sea bass (*Dicentrarchus labrax*) juveniles. *Aquaculture*, *205*(3–4), 287–299.

Polakof, S., Panserat, S., Soengas, J. L., & Moon, T. W. (2012). Glucose metabolism in fish: A review. *Journal of Comparative Physiology B: Biochemical, Systemic, and Environmental Physiology*, *182*(8), 1015–1045.

Pottinger, T. G. (1998). Changes in blood cortisol, glucose and lactate in carp retained in anglers' keepnets. *Journal of Fish Biology*, *53*, 728–742.

Rotllant, J., & Tort, L. (1997). Cortisol and glucose responses after acute stress by net handling in the sparid red porgy previously subjected to crowding stress. *Journal of Fish Biology*, *51*(1), 21–28.

Shimeno, S., Shikata, T., Hosokawa, H., Masumoto, T., & Kheyyali, D. (1997). Metabolic response to

feeding rates in common carp, *Cyprinus carpio*. *Aquaculture*, *151*(1–4), 371–377.

Sinha, A. K., Kumar, V., Makkar, H. P. S., De Boeck, G., & Becker, K. (2011). Non-starch polysaccharides and their role in fish nutrition - A review. *Food Chemistry*, *127*(4), 1409–1426.

Sulaiman, M.A., Kamarudin, M. S., & Ismail, M. F. S. (2017). The relationship between dietary carbohydrate level and the levels of blood glucose and amylase in lemon fin barb hybrid (*Barbonymus gonionotus* ♀ X *Hypsibarbus wetmorei* male ♂). *ATBU, Journal of Science, Technology & Education*, *5*(3), 110–119.

Waagbo, R., Glette, J., Sandnes, K., & Hemre, G. I. (1994). Influence of dietary carbohydrate on blood chemistry , immunity and disease resistance in Atlantic salmon , *Salmo salar L* . *Journal of Fish Disease*, *19*, 245–258.

