International Journal of Fauna and Biological Studies
Available online at www.faunajournal.com

IJFBS International Journal of Fauna And Biological Studies

ISSN 2347-2677
IJFBS 2018; 5(1): 245-247
Received: 28-11-2017
Accepted: 30-12-2017

Paloma D De Chavez
Marinduque State College, School of Fisheries-Gasan Campus, Banuyo, Gasan, Marinduque, Philippines.

Neil D Encinares
Technology Outreach Station for Marine Water Bureau of Fisheries and Aquatic Resources-Region 3, Bamban, Masinloc, Zambales, Philippines

Correspondence
Paloma D De Chavez
Marinduque State College, School of Fisheries-Gasan Campus, Banuyo, Gasan, Marinduque, Philippines.

Impact of *Aeromonas hydrophila* Infection on Freshwater Aquaculture Center Selected Tilapia (*Oreochromis niloticus*, FaST Strain)

Paloma D De Chavez and Neil D Encinares

Abstract
A study was initiated to evaluate the performance and hematological responses of Nile Tilapia FaST Strain *Oreochromis niloticus* (L) to an experimental infection with *Aeromonas hydrophila*.
The experiment consisted of two treatments with three replicates: Treatment 1 (Control)/non-injected fish (NI); Treatment 2 (Infected)/fish injected with *Aeromonas hydrophila*. Twenty-four hours after injection, the fish were anesthetized and the blood was collected. The hematological test included white blood cell (WBC) counts in treatments. Fish injected with *Aeromonas hydrophila* showed significant difference in white blood cell (WBC) counts than the non-injected treatment.
Final weight and weight gain increased significantly in fish injected when compared to non-injected control. Statistical analysis of survival rate showed significant difference between treatments ($P<0.05$). Treatment 1 (Control) has significantly higher survival rate which was 75% compared to Treatment 2 (Infected) which was 25%.

Keywords: Impact, *Aeromonas hydrophila*, Infection, Tilapia

1. Introduction
Nile Tilapia FaST Strain *Oreochromis niloticus* (L) is one of the recent strains of tilapia that are propagated at the Freshwater Aquaculture Center at Central Luzon State University, Science City of Munoz, Nueva Ecija, Philippines. Its propagation is well favored because of its rapid growth and the ability of this strain to utilize a wide variety of cheap feeds, both formulated and natural food organisms, including plankton, green leaves, benthic organisms, bacterial films, aquatic invertebrates and detritus (Teichert-Coddington, *et al.*, 1997) compared to other tilapia strains. This strain of tilapia has a number of special capabilities, including, filter feeding, efficient digestion, strong immune system, frequent breeding and mouth brooding. These can also be cultured in different aquatic environment in fresh, brackish and marine for it has been called "hardy" fish. Some of these capabilities occur in one fish or another, but seldom occur within the same fish as in the FaST strain. The fact that all of these characteristics occur within the same fish makes the FaST strains an ideal stock for aquaculture.
Bacterial diseases are among the most important causes of economic losses in cultured tilapia. *Aeromonas* spp., *Pseudomonas fluorescens*, *Vibrio anguillarum*, *Flavobacterium columnare*, *Edwardsiella tarda*, *Streptococcus* spp. and *Enterococcus* sp. are commonly found in aquaculture facilities (Plumb, 1997 [6]). They are often sub-clinical and without apparent signs. Under predisposing factors such as poor water quality, high ammonia as a result of high stocking density and feeding, ectoparasites, inadequate handling and stressful conditions, such microorganisms then found a portal of entry into the fish host (Moraes and Martins, 2004).
There are several studies on fish bacteria identification, experimental infection or disease resistance (Azad *et al.*, 2001; Al-Harbi [1] and Uddin, 2004; Cai, *et al*, 2004) but little relates the haematological parameters to bacterial experimental infection. This parameter is an important tool of diagnosis that reveals the state of health of fish (Blaxhall, 1972 [2]; Rehulka, 2002; Martins *et al.*, 2004a).
The profile of *Aeromonas* infection in this strain of Nile tilapia is not well documented. Clinical signs vary from sudden death in healthy fish to in-appetence, swimming abnormalities, pale gills, bloat and skin ulcerations. The skin ulcers may occur at any site on the fish and often they are surrounded by a bright red rim of tissue.

Because of the variability of these clinical signs, the diagnosis of this disease when based solely on the clinical presentation of the fish is highly unreliable and be economically disastrous to the fish producer.

The extent by which the hardy Nile Tilapia FaST Strain *Oreochromis niloticus* (L) react to infection with *Aeromonas hydrophila* is hereby reported in attempt to disseminate information to aquaculturists and fish farm owners.

2. Materials and Method

2.1 Experimental fishes

Ninety (90) pieces FaST Strain of Nile Tilapia (*Oreochromis niloticus*), fifty (50) grammers, were obtained from Freshwater Aquaculture Center. The experimental fish were randomly assigned to two treatments. Treatment 1 (Control, apparently healthy and no infection) and Treatment 2 (apparent signs of infection) consisting each of forty five (45) pieces of FaST Strain of Nile Tilapia (*Oreochromis niloticus*), fifty (50) grammers, with three (3) replicates; having fifteen (15) fishes each replicate. Experimental fishes were fed twice daily 9:00 AM and 3:00 PM and provided with good aeration. Monitoring for clinical signs, morbid responses and mortality was done daily.

2.2 Experimental Lay-out

Experimental fishes were all contained in aquaria with a dimension of 1.5 x 10^3 cfu/ml. FaST Strain of Nile Tilapia (*Oreochromis niloticus*) infected with *Aeromonas hydrophila* were also raised in aquaria with a pure culture of *Aeromonas hydrophila* previously isolated from an infected Genetically Male Tilapia (GMT) raised in ponds at the Freshwater Aquaculture Center (FAC), Central Luzon State University (CLSU), Munoz, Nueva Ecija, Philippines.

2.3 Determination of initial and final weight

Fish were weighed with the use of a digital weighing balance (Sartorius model) at the start and end of the experiment. Each treatment has an average initial weight of fifty (50) grams.

2.4 Determination of White Blood Cells (WBC) counts

Evaluation of White Blood Cell (WBC) profiles of fish in Treatments 1 and 2 was done at the start of the experiment and every week thereafter. Blood (100 µl) was collected intracardially with the use of a tuberculin syringe. A five (5) µl volume was added to 150 µl Turks solution (1:30 dilution). The blood and Turks solution suspension was placed on a hemocytometer slide (Burker-Turk, Tokyo) for WBC counting.

2.5 Statistical Analysis.

The experiment was conducted in conformity with Completely Randomized Statistical Design (CRD) with three replications. Mean responses were pooled as data for one treatment using One-Way Analysis of Variance (ANOVA). Treatment means were compared using Least Significant Difference (LSD).

3. Results and Discussion

Mean weight. Data on the initial weight, final weight and weight gain of FaST Strain of Nile Tilapia (*Oreochromis niloticus*) is shown in Table 1. The mean initial weights of fish did not vary in Treatment 1 and 2. After one month of feeding, the mean final weight of fish in Treatment 1 was 62.3g, which was significantly higher compared to mean final weight of fish in Treatment 2 (53.4 g).

Table 1: Weight gain, initial weight and final weight of uninfected and *Aeromonas hydrophila*- infected FaST Strain of Nile Tilapia (*Oreochromis niloticus*) during one month of rearing

Treatments	Initial Weight (g)	Final Weight (g)	Weight Gain (g)
T1 (Control)	50.0±	62.3±	12.3[a]
T2 (Infected)	50.0±	53.4±	3.4[b]

Means within a column with the same superscript are not significantly different at 5% probability level

Analysis of Variance (ANOVA) revealed significant difference in the computed weight gain for both treatments. Treatment 1 (Control) gained 12.3g which is significantly higher than 3.4 g for Treatment 2 (Infected).

Mean WBC Count White Blood Cell (WBC) data of FaST Strain of Nile Tilapia (*Oreochromis niloticus*) during the experimental period is shown in Table 2. Statistical analysis showed no significant difference in the initial White Blood Count (WBC) profile between treatments. Initial count was 2.77 x 10^7 and 2.77 x 10^7 for Treatment 1 and 2 respectively.

Table 2: White Blood Cell (WBC) profile of uninfected and *Aeromonas hydrophila*- infected FaST Strain of Nile Tilapia (*Oreochromis niloticus*) during one month of rearing

Treatments	Initial Count	1st Week	2nd Week	3rd Week	4th Week
T1 (Control)	2.77 x 10^{7a}	3.15 x 10^{7a}	3.24 x 10^{7a}	3.20 x 10^{7a}	3.22 x 10^{7a}
T2 (Infected)	2.77 x 10^{8a}	5.54 x 10^{8b}	7.34 x 10^{8a}	2.3 x 10^{9a}	3.3 x 10^{9b}

Means within a column with the same superscript are not significantly different at 5% probability level

Statistical analysis revealed significant difference in WBC profile of FaST Strain of Nile Tilapia (*Oreochromis niloticus*) in the 1st week of observation between treatments. Treatment 2 (Infected) had significantly higher WBC Count compared to Treatment 1 (Control). However, in the 2nd and 3rd week WBC count reveals no significant difference observed in the Treatments. There was significant difference in the WBC count in the 4th week of observation.

Survival Rate The survival rate of FaST Strain of Nile Tilapia (*Oreochromis niloticus*) after one month of rearing is presented in Table 3. Statistical analysis revealed significant difference in the survival rate between treatments. Higher survival rate was obtained in Treatment 1 (Control) compared to that of Treatment 2 (Infected).

Table 3: Survival rate of uninfected and *Aeromonas hydrophila*- infected FaST Strain of Nile Tilapia (*Oreochromis niloticus*) after one month of rearing

Treatments	Survival Rate (%)
T1 (Control)	75%[a]
T2 (Infected)	25%[b]

Means within a column with the same superscript are not significantly different at 5% probability level.

Tilapia is one of the most cultivated freshwater fish worldwide. This study was designed to determine if injection

with *Aeromonas hydrophila* bacteria isolated from diseased tilapia is responsible for haematological changes. The variation degree on the haematological response is an important tool to fish health diagnosis and may vary according to stressor stimulus, treatment, parasitic or infectious diseases (Silveira-Coffigny *et al*., 2004 [8]; Chen *et al*., 2004; Martins *et al*., 2004a; Rehulka, 2002).

In terms of growth parameter, results of this experiment revealed that better growth was attained in Treatment 1 (Control). As reviewed by Frazier (1988), *Aeromonas hydrophila* is pathogenic to fish which causes detrimental effects in the physiological and biological state of the species. Furthermore, it is expected that fish subjected to any infection would greatly contribute to fish's health making them more vulnerable, thus weakens their immunity to diseases.

Fish injected with *Aeromonas hydrophila* were found to have the higher values of white blood cell (WBC) counts in the differential counting besides a decreased number of monocytes. In fact, under severe infection the organism produces more white blood cells. It can be added that lymphocytes have been reported as immunocompetent cells (Ellis *et al*., 1976 [3]).

Contrarily, Rafiq *et al* (2001 [7]) did not observe any alteration in the differential counts of white blood cells in tilapia infected with *A. hydrophila*. In carp experimentally infected with *A. hydrophila*, Harikrishnan *et al.* (2003) have related increased WBC counts, confirming these results. According to this author, decreased RBC counts and hematocrit indicate that erythrocytes are being affected or destroyed with the infection.

The low survival rate of injected fish compared to non-injected could be attributed to the *Aeromonas hydrophila* infection. It was also observed that they produced more fecal wastes which in turn, make dissolved oxygen lower.

4. Conclusion and Recommendation

The experiment was conducted to evaluate the Impact of *Aeromonas hydrophila* Infection on Freshwater Aquaculture Center Selected Tilapia (*Oreochromis niloticus*, FaST Strain). Two Treatments were used in the experiment wherein Treatment 1 is the control group without infection and Treatment 2 was the infected group.

Based on the results of the study, higher weight gain and survival rate was observed in Treatment 1 (Control) compared to that of Treatment 2 (Infected).

The findings showed that FaST Strain of Nile Tilapia (*Oreochromis niloticus*) was hardly affected with *Aeromonas hydrophila* infection. Therefore, immunity of FaST Strain of Nile Tilapia (*Oreochromis niloticus*) is greatly facilitated on how the infection was being introduced.

It is hereby recommended that studies on the different strains of tilapia be conducted to evaluate its comparable effect in different parameters. Furthermore, study on different confinement should be done to determine the effects on different environments.

5. References

1. Al-Harbi A, Uddin MN. Seasonal variation in the intestinal bacterial flora of hybrid tilapia (*Oreochromis niloticus* x *Oreochromis aureus*) cultured in earthen ponds in Saudi Arabia *Aquaculture* 2004; 229(1-4):37-44.
2. Blaxhall PC. The haematological assessment of the health of freshwater fish. A review of selected literature J Fish Biol 1972; 4(4):593-604.
3. Ellis A.E, Munro Als, Roberts RJ. Defense mechanisms in fish. A study of the phagocytic system and the rate of intraperitoneally injected particulate material in the place (*Pleuronectes platessa* L) *J Fish Biol*. 1976; 8(1):67-78.
4. Hamilton A. "DNA relationships and transfer to antibiotic resistance among motile Aeromonads from human, fish and environmental specimens, Aquaculture News, Institute of Aquaculture, University of Stirling, Scotland, 1996, 37.
5. Krishna R.R, K.G Rao, P.H Babu. White Spot Disease, World Aquaculture. World Aquaculture Society USA, 1997, 25-37.
6. Plumb JA. Infectious diseases of tilapia. In Costa-Pierce, BA and RAKOCY, JE (Eds) *Tilapia aquaculture in the Americas* Baton Rouge, Luisiana, USA: World Aquaculture Society, 1997, 212-228.
7. Rafiq M, Sarder I, Thompson KD, Penman DJ, Mcandrew BJ. Immune responses of Nile tilapia (*Oreochromis niloticus* L) clones: I Non-specific responses *Dev Comp Immunol* 2001; 25(1):37-46.
8. Silveira-Coffigny R, Prieto-Trujillo A, Ascencio-Valle F. Effects of different stressors in haematological variables in cultured *Oreochromis aureus* S Comp Biochem Physiol C 2004; 139(4):245-250.
9. Swann L. Diagnosis and Treatment of "Aeromonas hydrophila" Infection of Fish Animal Disease Diagnostic Laboratory. Purdue University Illinois, 2000.
10. Tonguthai K, S Chinabut. Diseases of Tilapia. *In:* H.S Egna and C.E Boyd (EDS) Dynamics of Pond Aquaculture CRC Press LLC, 1997, 42-56.