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Effect of dietary salt supplementation on growth and feed utilization of Tilapia (*Oreochromis niloticus*)

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Abstract
The present study was conducted to determine the consequences of supplementing dietary salt on growth and feed utilization of tilapia (*Oreochromis niloticus* Linnaeus, 1758) in laboratory condition for a period of 21 days. Four treatments with three replicates in each containing 30 fish (average weight, 2.2 ± 0.01 g) were stocked in 100 liter aquaria. Four treatments were designated as T_1 (SD+0% salt of diet), T_2 (SD+1.0% salt of diet), T_3 (SD+1.5% salt of diet) and T_4 (SD+2% salt). The fish were fed at 20% body weight twice a day. Significantly ($p<0.05$) higher weight gain of tilapia was found in T_3 (8.90 ± 0.31g) followed by T_2 (8.75 ± 0.29 g), T_4 (7.68 ± 0.07 g) and T_1 (5.23 ± 0.22 g). The highest length gained by the groups treated with T_3 (2.93 ± 0.19 cm) followed by T_2 (2.92 ± 0.04 cm), T_4 (2.73 ± 0.06 cm) and T_1 (1.84 ± 0.03 cm). Specific growth rate (SGR) ($p<0.05$) was found to be the highest in T_3 (8.16 ± 0.21) followed by T_2 (8.08 ± 0.23), T_4 (7.56 ± 0.06) and T_1 (6.20 ± 0.24). The highest survival was found in T_3 (85.55 ± 1.93) followed by T_2 (84.44 ± 1.93), T_1 (83.33 ± 0.00%) and T_4 (81.11 ± 1.92). The lowest food conversion ratio (FCR) was obtained in T_2 (1.15) than that of T_1 (1.57), T_4 (1.40) and T_3 (1.18), respectively. The growth parameters indicated that supplemental feed with 1.5% dietary salt can be incorporated in *O. niloticus* diet.

Keywords: *Oreochromis niloticus*, dietary salt, supplementation, growth, feed utilization

1. Introduction
Fish is the second most valuable agricultural crop in Bangladesh and its production contributes to the livelihoods and employment of millions of people. Fisheries in Bangladesh are diversified; there are about 795 native species of fish and shrimp in the fresh and marine waters of Bangladesh and 12 exotic species that have also been introduced [1]. In 2014-15, the total fish production is 3.684 million tonnes of which 55.93% come from aquaculture [2]. Inland open, inland closed and marine waterbodies represent 1023991, 2060408 and 599846 tonnes, respectively [2]. In the agro-based economy of Bangladesh, Fisheries sector contributed 3.69% to national GDP [2]. Among exotic carps, tilapia is one of the promising species which is being cultured throughout the country. Tilapia culture is also one of the fastest growing farming activities, with an average annual production of 347801 tonnes [3]. In view of the increasing commercialization and continuing growth of tilapia industry, the commodity is not only the second most important farmed fish globally, next to carps but also described as the most important aquaculture species of the 2lst century [4]. As the human population continues to grow, finding means to feed those people is one of the most important challenges faced around the globe. Bangladesh is brought face to face today with the problem of protein calorie malnutrition. To overcome these problems of protein malnutrition increase of fish production is essential. Therefore, a significant effort to increase fish production should be concentrated on aquaculture. Tilapia is one of the most important groups of fish for aquaculture and contributes 9.44% to the total production of fish in Bangladesh [3]. As tilapia culture continues to expand worldwide, there is a growing pressure to minimize production cost associated with diet supplementation. Supplementation of diets with growth inducing substances has the potential to be profitable because of the improved growth rate or reduced culture period. Additives like sodium chloride are essentially ideal to enhance growth if incorporated in artificial feed as supplements. The use of salt (NaCl) is not a new advent. Salt is one of the essential mineral elements required by the animal and plant bodies for their normal functioning namely; making food taste better, regulating osmotic pressure of the body, form acid in mucous membrane of the stomach (activation of pepsin and enzymes of the salivary glands of

the throat and keeping digestive processes normal). According to Nasir and Hamed (2016) [6] dietary supplementation of NaCl influenced the body composition and can be beneficial for the young common carp reared in freshwater used in the fish culture. Mzengereza *et al*. (2015) [7] found that 1.5% salt in fish diets enhance growth and better feed conversion ratio.
Considering the above facts, the study was conducted with the following objectives to know the effects of dietary salt supplementation on growth and survival of *Oreochromis niloticus* and to assess the optimal level of salt to be incorporated as a supplement in fish diet.

2. Materials and Methods

2.1 Study Area

The experiment was conducted inside the Post-graduate laboratory of the Department of Fish Biology and Genetics, Faculty of Fisheries, Sylhet Agricultural University, Sylhet. The experiment was carried out for a period of 21 days from 18 May to 7 June 2016 in 12 aquariums.

2.2 Collection and acclimatization of tilapia fry

Healthy and uniformed sized Tilapia (*Oreochromis niloticus)* fry were collected from a private hatchery (Pirojpur Fishery Khamar, Dakhsin Surma, Sylhet) and kept into an oxygenated polybag. Before stocking fry were acclimatized to the aquarium water gradually for one hour.

2.3 Experimental design

The experiment was laid out in a Completely Randomized Design (CRD) using 100 liters aquarium each stocked with 30 fry. Four treatments with three replicates for each were used for the experiment. The treatments were designated as T_1 (control), T_2, T_3 and T_4. T_1 contains 0%, T_2 contains 1.0%, T_3 contains 1.5% and T_4 contains 2.0% dietary salt in diet, respectively. The design of experiment is shown in Table 1 and the ingredient of feed used in the experiment is shown in Table 2.

Table 1: Experimental design of the research work

No. of Treatment	Salt supplementation in diet (%)	Feed	No. of Replication	Water volume in each tank (liter)	Stocking density (No./treatment)
T_1(control)	-	*SD	3	100	30
T_2	1.0%	*SD	3	100	30
T_3	1.5%	*SD	3	100	30
T_4	2.0%	*SD	3	100	30

Picture 1: Layout of experimental unit

Table 2: The ingredient of feed used in the experiment

Treatments	Feed ingredients
T_1	20% SD of fish body weight
T_2	20% SD of fish body weight + 1.0% salt supplementation of feed weight
T_3	20% SD of fish body weight + 1.5% salt supplementation of feed weight
T_4	20% SD of fish body weight + 2.0% salt supplementation of feed weight

*SD = Supplemental Diet

2.4 Supplemented diet

The stocked fry of tilapia were fed with commercial starter feed (containing 32% protein) at the rate of 20-10% of body weight twice daily (Table 3).

Table 3: Proximate composition of supplementary feed used in the experiment

Major composition of diet	Amount of composition (%)
Improved protein % (lowest)	32
Moisture % (highest)	11
Crude lipid % (lowest)	5
Crude fiber % (highest)	5

2.5 Experimental Procedure
Tilapia fries were collected from a commercial hatchery with initial mean weight of 2.2±0.01 g and initial mean length of 5.0±0.01 cm and kept in 100 liter rectangular aquarium for 2 hours acclimatization. After acclimatization weight and length were recorded and fries were stocked in aquarium according to the design of the experiment (Table 1).

The aquarium was provided with sand and iron filtered freshwater (2 $lmin^{-1}$) and with continuous aeration to maintain the dissolved oxygen level near saturation. Tilapia were fed twice daily at 09.00 am and 4.00 pm. Fish were weighed in bulk every week and measured the length until the ending of the experiment (3 weeks). Fecal matters were removed by siphoning at each morning before morning feeding and at each afternoon before afternoon feeding and half of the water replaced with the filtered water twice daily after the removal of the fecal matters and before the feeding. Mortality was cheeked daily. The experiment was conducted at ambient temperature with a natural photoperiod (approximately 12 h light and 12 h dark). Water temperature and salinity were monitored daily between 08.00 to 09.00 am, while pH and dissolved oxygen were monitored twice weekly.

Every 7 days intervals in the morning (9.00 to 10.00 am), approximately 15 h after the last feeding all fish were counted and randomly selected 6 fishes were weighted from each replication to determine weight gain, specific growth rate and length increment. Weight was taken with a spring balance (DONGIL-15 kg x 50 g) and length was measured with a measuring scale. All the data were recorded in a note book and finally calculated the average length and weight of fishes according to treatment on each sampling day. After taking the data, all the fishes were kept to their own aquarium carefully. At the ending of the 3 weeks experiment, all fishes were counted and individually weighted to determine the survival, weight and length.

The following formulae were used to evaluate the growth:

a) Length gained = Mean final length – Mean initial length

b) Weight gained = Mean final weight – Mean initial weight

c) $$\text{Survival rate (\%)} = \frac{\text{No. of fish caught}}{\text{No. of Fish released}} \times 100$$

d) Specific growth rate:

SGR (% /day) = [(ln W_2 − ln W_1) /duration] × 100

Where, W_1 = Initial live body weight (g) at time T_1 (day)

W_2 = Final live body weight (g) at time T_2 (day)

e) $$\text{Feed conversion ratio (FCR)} = \frac{\text{Amount of feed distributed}}{\text{Weight gain of fish}}$$

2.6 Statistical analyses
Growth, survival and production parameters were analyzed using one way analysis of variance (ANOVA) to compare the treatments means. If the main effect was found significant, the ANOVA was followed by Duncan's Multiple Range Test (DMRT) at 5% level of significance using SPSS version 22.

3. Results and Discussion
3.1 Weight
Mean body weight of tilapia fry during stocking was 2.2 g in all aquariums. Growth of tilapia fry in all treatments increased throughout the rearing period. Significantly ($p<0.05$) higher mean final weight was recorded in T_3 (11.07±0.29 g) followed by T_2 (10.92±0.25 g) and T_4 (9.86±0.11 g) while the lowest in the control diet T_1 (7.49±0.17 g).

Table 4: Growth (Mean ± SD) of tilapia in aquarium after 21 days of the experiment

Parameters	Treatments			
	T_1	T_2	T_3	T_4
Stocking density	30.00	30.00	30.00	30.00
Average initial weight (g)	2.2 ± 0.06	2.2 ± 0.05	2.2 ± 0.06	2.2 ± 0.05
Average final weight (g)	7.49 ± 0.17^a	10.92 ± 0.25^c	11.07 ± 0.29^c	9.86 ± 0.11^b
Mean final weight gain (g)	5.23 ± 0.22^a	8.75 ± 0.29^c	8.90 ± 0.31^c	7.68 ± $0..07^b$
% of weight gain	245.98 ± 16.71^a	403.58 ± 23.05^c	411.38 ± 21.33^c	353.76 ± 5.88^b
SGR	6.20 ± 0.24^a	8.08 ± 0.23^c	8.16^c ± 0.21^c	7.56 ± 0.06^b

Different superscript letter in the same raw indicated significant differences ($p<0.05$)

Mean weight gain and % weight gain of tilapia (*O. niloticus*) varied significantly among the four treatments. The lowest weight gain and % weight gain (5.23±0.22 & 245.98±16.71) were observed in fish fed Diet 1, which was significantly lower than WG observed for fish fed Diet 2, 3 and 4. The highest weight gain and % weight gain (8.90±0.31 & 411.38±21.33) were found in T_3 which were significantly higher (p<0.05) than those of T_1 and T_4 but there is no significant difference between T_3 and T_2.

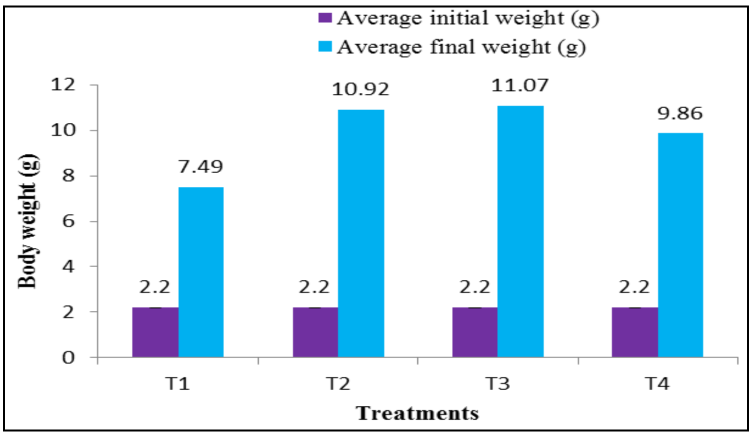

Fig 1: Growth rate of tilapia (*Orechromis niloticus*) under different treatments

Significantly ($p<0.05$) highest final weight of tilapia was found in T_3 (11.07±0.29 g) and T_2 (10.92±0.25) than those of T_4 (9.86±0.11) and T_1 (7.49±0.17 g). Results showed that higher fish growth was observed at 1% and 1.5% salt inclusion level (Table 4.1). Thus, consistent to a large dimension with other researchers in that significant growth can be observed when fish are fed on salty diet than salt free diet. Keshavanath *et al.* (2011) [5] indicated that the final weight of Rohu, *Labeo rotiha* (62.50 g) fed 0.5% and (75.69 g) fed 1% NaCl–incorporated diets were significantly ($p<0.005$) higher than that of the control (51.12 g) 0% NaCl. The addition of salt to the diet of common carp at a level of 1.5% resulted in significantly better growth (33.85±0.49 g) with initial mean weight of 20.50±0.28 g and significantly lower values were found in fish fed control diet 26.10±0.42 g [6]. Mzengereza *et al.* (2015) [7] observed that higher growth of *O. shiranus* was at 1.5% (18.69±0.22) and 1% (17.19±0.19) than control diet (16.32±0.99). Keshavanath *et al.* (2012) [8] reported that final mean weight of Tambaqui was higher at 2% dietary salt (4.77±0.22) and 1.5% (4.76±0.16). Cnaani *et al*. (2012) [9] found that the highest growth of white grouper was 19.2±0.5 at 3% dietary salt supplementation than the control diet (13.2±1.2). The highest growth of *Labeo rohita* was 9.12±1.48 at 2% NaCl incorporated diet and the lowest growth of Rohu was 6.08±1.43 at control diet [10]. Copatti *et al.* (2011) [11] reported that final mean weight of silver catfish was 5.83±0.35 at 1% NaCl containing diet than control diet (6.67±0.22). Dietary salt has been found to improve fish growth and feed conversion efficiency in several species: the Japanese eel [12], rainbow trout [13], common carp and mrigal [14], rohu [15] and gilthead sea bream [16]. In an earlier study, it was found that dietary NaCl induced growth in common carp and mrigal [14].

Freshwater fish, being osmotic to the surrounding medium encounter the physiological problem of solute loss and in order to compensate this, they resort to active uptake of salt ions from the medium. Therefore, it is reasonable to speculate that dietary salt satisfied the osmolegulatory requirement of *O. niloticus* which is a freshwater fish and that supplemental salt spared energy used in osmoregulation, thereby leaving more energy for somatic growth. Smith *et al.* (1989) [17] reported that the dietary sodium intake of salmonids kept in fresh water increased by eightfold from winter to summer. This corresponds to the increase in feeding and shows that almost all the sodium required can be derived from dietary salt. This can therefore be used as a source of salts for fish kept in freshwater, providing ions which the fish cannot obtain from the hypotonic environment.

4.2 Specific growth rate (SGR)

The mean specific growth rates (SGR) of tilapia in different treatments were 6.20±0.24, 8.08±0.23, 8.16±0.21 and 7.56±0.06 in T_1, T_2, T_3 and T_4, respectively. Significantly ($p<0.05$) highest SGR value (8.16±0.21) was found in T_3 whereas the lowest SGR value (6.20±0.24) was found in T_1 (Table 4 and Fig. 2).

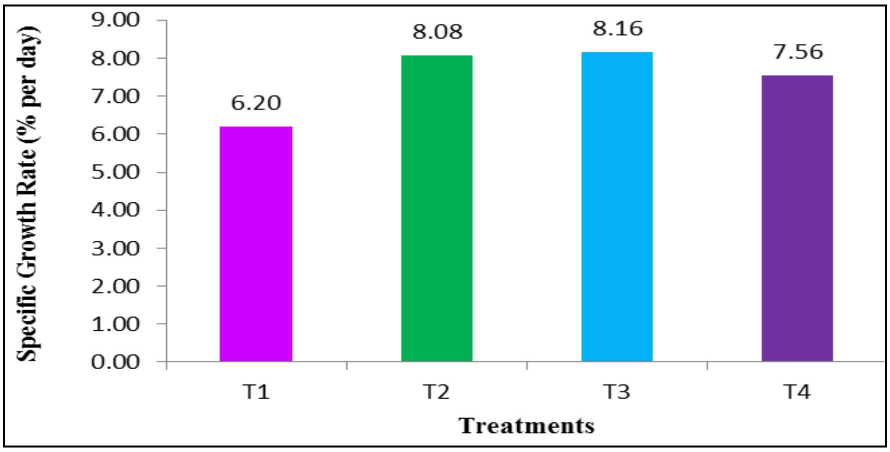

Fig 2: Specific growth rate of tilapia (*Orechromis niloticus*) under different treatments

SGR of Tambaqui at 2% salt is 2.47±0.07 [8]. Mzengereza *et al.* (2015) [7] found that SGR of *O. shiranus* was 0.52±0.03 at 1.5% NaCl. Gangadhara *et al.* (2014) [10] reported that 2% NaCI produced highest SGR 5.45±0.16 than control diet 5.04±0.24 in *Labeo rohita*. Eroldogan *et al.* (2005) [18] reported that SGR of European sea bass was 2.3±0.1 at 3% NaCl. Keshavanath *et al.* (2011) [5] reported that 2% NaCI produced the highest SGR 2.97±0.14 than control diet 2.67±0.21. Highest SGR of *Labeo rohita* was 2.22±0.03 fed 1% NaCl than the control diet 1.90±0.03 [10].

4.3 Length

The mean final length of tilapia (*O. niloticus*) varied among the four treatments. The highest length was found in T_3 (7.97±0.25) and the lowest length was found in T_1 (6.88±0.08). Tilapia fry fed with the control diet (Diet 1) without salt showed the lowest length gain in this experiment. T_3 (7.97±0.25) which contained 1.5% dietary salt showed the highest length gain.

Table 5: Length (Mean ± SD) of tilapia in aquarium during the experimental period

Parameters	Treatments			
	T_1	T_2	T_3	T_4
Average initial length (cm)	5.00 ± 0.06	5.00 ± 0.06	5.00 ± 0.06	5.00 ± 0.06
Average final length (cm)	6.88 ± 0.08^a	7.95 ± 0.07^c	7.97 ± 0.25^c	7.77 ± 0.02^b
Mean final length gain(cm)	1.84 ± 0.03^a	2.92 ± 0.04^c	2.93 ± 0.19^c	2.73 ± 0.06^b
% of length gain	36.62 ± 0.40^a	57.95 ± 0.93^b	58.28 ± 1.93^b	54.28 ± 1.93^b

Different superscript letter in the same raw indicated significant differences ($p<0.05$)

The mean length gains of tilapia (*O. niloticus*) were 1.84±0.03, 2.92±0.04, 2.93±0.19 and 2.73± 0.06 in T_1, T_2, T_3 and T_4, respectively (Table 5).

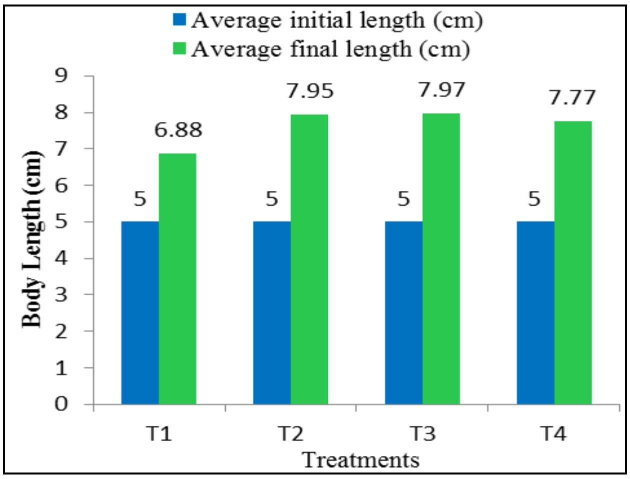

Fig 3: Variation of body length (cm) of tilapia under different treatments

The mean final lengths of tilapia (*O. niloticus*) were 6.88±0.08 cm, 7.95±0.07 cm, 7.97±0.25 cm and 7.77±0.00 cm in T_1, T_2, T_3 and T_4, respectively after 21 days experiment with 0%, 1%, 1.5% and 2% dietary salt of supplemental diet, respectively. The highest length was recorded in T_3 (7.97±0.25) and the lowest length in T_1 (6.88±0.08). This result supported by the previous work done by other scientists with dietary salt. Keshavanath *et al.* (2012) [8] reported that final mean length of Tambaqui higher at 2% dietary salt (6.79±0.16) and 0.5% (6.54±0.25) than control diet (6.19±0.19). The highest mean length of *Labeo rohita* was 9.10±1.23 at 1% NaCl incorporated diet than the control diet [10]. Copatti *et al.* (2011) [11] reported that final mean length of silver catfish was 8.57±0.27 cm at 1% NaCl containing diet than control diet (7.89±0.16 cm).

4.4 Food conversion ratio (FCR)

Food conversion ratio (FCR) of the present investigation was ranged between 1.15-1.57. Feed conversion ratios (FCR) of tilapia in four treatments were 1.57±0.09, 1.15±0.02, 1.18±0.06 and 1.40±0.02 in T_1, T_2, T_3 and T_4, respectively. FCR was significantly ($p<0.05$) lower in T_2 (1.15) than that of T_1 (1.57) and T_4 (1.40) but T_3 (1.18) was not significantly differ from T_2.

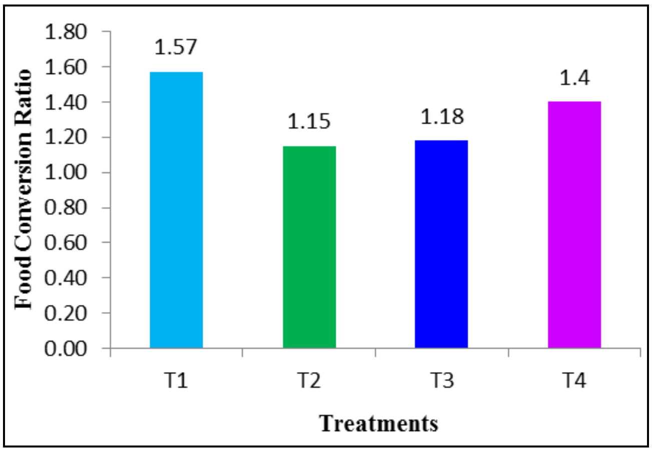

Fig. 4: Food conversion ratio of tilapia (*O. niloticus*) under different treatments

Gangadhara *et al.* (20014) [10] reported that 0.5% NaCl and 1% NaCl produced better FCRs at 1.56 and 1.44, respectively and subsequently higher growth rate. Keshavanath *et al.* (2011) [5] reported that adding 1% salt to the diet of (*Labeo rohita*) resulted in significant improvement in their feed conversion ratio 1.47 than control diet 1.77. Mzengereza *et al.* (2015) [7] observed that *O. shiranus* exhibited a better feed conversion ratio at 1.5% NaCl diet 1.44±0.08. Tambaqui fed 1.5% salt recorded best FCR 2.02±0.13 than control diet 2.35±0.12 [8]. Feed conversion ratio for common carp is better by feeding diets with 1.5% (2.21±0.007) added salt [6]. Fontainhas-Fernandes *et al.* (2000) [19] reported that adding salt to fish diet increases appetite and digestibility. Gatlin *et al.* (1992) [20] reported that juvenile red drum exhibited greater feed efficiency and significant weight gain when 2% NaCl was added to their diet. Eroldogan *et al.* (2005) [18] found that 5% salt supplementation in the diet of European sea bass enhanced growth and feed utilization.

Fish feeds constitute one of the most expensive components in rearing of fish, and high protein levels required for these fish are also a major source of nitrogenous products harmful to fish in closed systems. Therefore, the fact that the addition of salt to the feed resulted in a better FCR is of great importance as this is an indication of good feed conversion into fish. 2% NaCl had the highest FCR clearly showing that feed utilization was poor. Low digestibility and faster evaluation of food have been associated with high levels of NaCl in diets. This in turn could affect assimilation and conversion efficiency. It is, therefore, reasonable to suggest that 2% salt level led to excessive salt loading and adversely affected feed intake.

4.5 Survival

The survival of tilapia was ranged from 81.11 to 85.55%. The survival in T_1, T_2, T_3 and T_4, were 83.33±0.00, 84.44±1.93, 85.55±1.93 and 81.11±1.92, respectively. Higher survival of tilapia was found in T_3 (85.55%) and T_2 (84.44%) while lower in T_4 (81.11%). However highest survival was observed in T_3 where 1.5% salt supplementation feed added.

Table 6: Survival rate of tilapia (*O. niloticus*) in aquarium after 21 days of experiment

Treatments	Survival rate (%)
T_1	83.33 ± 0.00
T_2	84.44 ± 1.93
T_3	85.55 ± 1.93
T_4	81.11 ± 1.92

Keshavanath *et al.* (2011) [5] reported that adding 1.5% salt to the diet of (*Labeo rohita*) resulted survival of 88%. Mzengereza *et al.* (2015) [7] found that survival of *O. shiranus* was 94.8±0.8 at 1% dietary salt supplementation. Cnaani *et al.* (2012) [9] reported that highest survival of white grouper was 77.8±3.9 at 3% dietary salt supplementation than the control diet (60.7±2.5). The highest survival of *Labeo rohita* was 88% at 1.5% and 84% at 1% NaCl incorporated diet reported by Gangadhara *et al.* (2014), which was more or less similar of the present study [10].

4. Conclusion and Recommendations

The present study reveals that supplementation of dietary salt with feed play a significant role in increasing the growth of tilapia fry. In respect of tilapia weight, length, SGR and survival rate, T_3 treatment performed the best growth performance. Salt level of 1.5% (T_3) is optimum for incorporation in diets of *Oreochromis niloticus* where 1% inclusion level plays second fiddle. Feed utilization was better in both 1% and 1.5% salt inclusion levels with 1.15 and 1.18 FCRs, respectively. At 2% salt inclusion the SGR, FCR, survival and weight gain diminished, thus an increase in salt level beyond a certain (optimum) level make the feed less appropriate for consumption and affect growth negatively. However, on the basis of information obtained from this study, it is concluded that the supplementation of 1.5% dietary salt with feed may be recommended for successful *O. niloticus* culture. The further study should be carried out in different agro-ecological zones to standardize and refinement of the technology.

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