

NORWEGIAN UNIVERSITY OF LIFE SCIENCES



Abstract

Chowdhury, D.K. Optimal feeding rate for Nile tilapia (*Oreochromis niloticus*). MSc thesis. Department of Animal and Aquacultural Sciences, Norwegian University of Life Sciences, Submitted 2011-05-16.

The aim of this study was to define optimal feeding rates for Nile tilapia (*Oreochromis niloticus*). Four experiments were carried out to evaluate the effect of feeding rate on growth performance of larger and juvenile tilapia by means of estimating growth rates, apparent nutrient digestibilities, feed utilization, body compositions, and nutrient and energy retentions. One nutritionally balanced diet (crude protein 342, crude fat 67, ash 47, starch 251 (all values in g (kg dry matter)⁻¹)) was prepared by extrusion and used in all experiments. The experiments were carried out in a freshwater recirculation system. The temperature was maintained at 25 to 26°C, pH at 6.8 and oxygen above 5 mg l⁻¹. Duplicate tanks were used for each feeding rate treatment.

The first experiment was carried out with duration at six weeks, and utilized adult tilapia (258 ±0.3 g, mean±SEM). Fishes were fed six times in 24 hr photoperiod at five feeding rates (55, 70, 85, 100 and 115% where 2.25% body weight per day as 100% satiation). Daily weight gain was predicted assuming that the feed conversion ratio (FCR) was 1.1 g feed dry matter (DM) intake per g gain. Analysis of variance showed that 55% feeding rate is like to be proficient for FCR at 1.01. Energy content of tilapia was significantly higher at 100-115% feeding rate and intermediate at 55 to 85% feeding rate. Nitrogen retention was significantly higher at limited feeding rate (55%). Apparent crude protein digestibility was found significantly higher at 100% satiation and intermediate at 70, 85 and 115% feeding rate. The estimated optimal feeding rate was at the 55% level, corresponding to 1.2 % a day.

The second experiment was conducted over a 4 week period with juvenile tilapia (1.1±0.02 g) fed 22 times in 24 hr photoperiod at five feeding rates (55, 70, 85, 100 and 115% where 8 and 6% body weight per day considered as 100% satiation for 1st and 2nd two week, respectively). FCR was set at 0.8 g DM intake per g gain. Growth rates were significantly higher at 85-115% feeding rate while FCR (0.66 to 0.81) significantly lower at 55-85% feeding rate. Whole-body dry matter, crude protein, crude fat, and energy contents were significantly elevated for the highest feeding rates, while ash was reduced. Nitrogen, phosphorus and energy retentions were significantly

lowered at restricted feeding. The estimated optimal feeding rate based on growth rates was 6.8% a day for the first 2 weeks and 5.1% for the next two weeks.

The third experiment was designed with 4 weeks feeding of juvenile tilapia (1.1 ± 0.02 g) in accordance with same frequency and photoperiod as same as second experiment. Fishes were fed at four declining and one fixed feeding rate 8-16 to 8 and 6-14 to 6% body weight for 1st and 2nd two week respectively. Weight gain% and SGR were significantly higher at 10-8 and 8-6 to 16-8 and 14-6% feeding rate while FCR (0.66 to 0.71) were significantly lower at 10-8 to 8 and 8-6 to 6% feeding rate. Dry matter, crude protein, crude fat, ash and energy content were significantly affected by declining feeding rate and higher than initial body composition. Nitrogen and total phosphorus retentions were significantly higher at lesser feeding regime while energy retention was significantly higher at 12-8 and 10-6% feeding rate. The estimated optimal feeding rate based on growth rates was to reduce feeding from 10 to 8% a day for the first 2 weeks and from 8 to 6% for the next two weeks.

The fourth experiment was carried out at two week duration for larger tilapia (77.9 ± 0.03 g). Fishes were fed at five feeding rate (1, 2, 3, 4 and 5% body weight a day) with same frequency and photoperiod as experiment 2 and 3. FCR was set at 0.9 g DM intake per g gain. Growth rates were significantly higher at 3 to 5% feeding rate while FCR (0.86 to 0.89) were significantly lower at 1 to 3% feeding rate. Dry matter and energy content were significantly increased by feeding rate but ash content was significantly decreased as feeding rate increase. Nitrogen retention was significantly higher at 1-3% feeding rate while energy retention at 3% feeding rate. The estimated optimal feeding rate based on growth rates was 3% a day.

In conclusion, correct feeding rate can be used for maximize growth and feed utilization for genetically improved Nile tilapia. Declining feeding rate is better than fixed feeding rate and can be suitable for Juvenile tilapia (1.1 g) at 10-8% and 8-6% for 1st and 2nd two week, respectively while 3% feeding rate can be proper for tilapia between 80 and 115 g. Tilapia larger than 260 g likely to be proficient at 1.2% body weight.

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List of abbreviation and symbols

ANOVA	Statistical tool, Analysis of Variance
DM	Dry matter
Expt	Experiment
FAO	Food and Agricultural Organization
FM	Fish meal
GIFT	Genetically Improved Farmed Tilapia
GST	Genetically Supreme Tilapia
ln	Natural logarithm
MMT	Million metric ton
NO ₂	Nitrite
S.E.M.	Standard error of mean
SGR	Specific growth rate
TAN	Total ammonia nitrogen
UMB	Norwegian University of Life Sciences
WG	Weight gain

CHAPTER ONE

1: Introduction:

Tilapia (*Oreochromis spp*) are now commercially important fish and grown in almost 100 countries. They have become among the most important food fishes in the world (Lim and Webster, 2006). Global production of farmed Nile tilapia was 1.66 million metric ton (MMT) and 2.54 MMT in 2005 and 2009, respectively (Fig.1). Including other cichlids the production was 3.1 MMT out of global aquaculture production of 55.1 MMT (FAO, 2010). Thus tilapia and other cichlids totally contribute about 5.6% of total aquaculture production.

So it appears that tilapias are likely to be higher rank in global aquaculture production next to carp production. According to El-Sayed (2006) the attributes that makes tilapia as an ideal candidate for aquaculture, especially in developing countries are:

- Rapid growth,
- Omnivorous fish, can use high proportion of inexpensive plant sources in their feeds,
- Stands well in wide range of environmental conditions (Such as temperature, salinity, low dissolve oxygen, etc.).
- Resistance against stress and diseases.
- Short generation interval and
- Low supplementary feed require in natural environment and can take the commercial feed immediately after yolk-sac absorption,

Intensification of tilapia farming has been promoted and farmers are enhancing growing condition of fish (Asche et al., 2008). The authors also mentioned that innovation of production technology to exploit the biological merits of tilapia has played important role to up lift the farming and the production as well. At the same time, Fig.2 shows that tilapias' are distributed globally and it has consumers all over the world, requires minimum fish meal, suitable for different culture system either in marine or fresh water (Fitzsimmons, 2010).

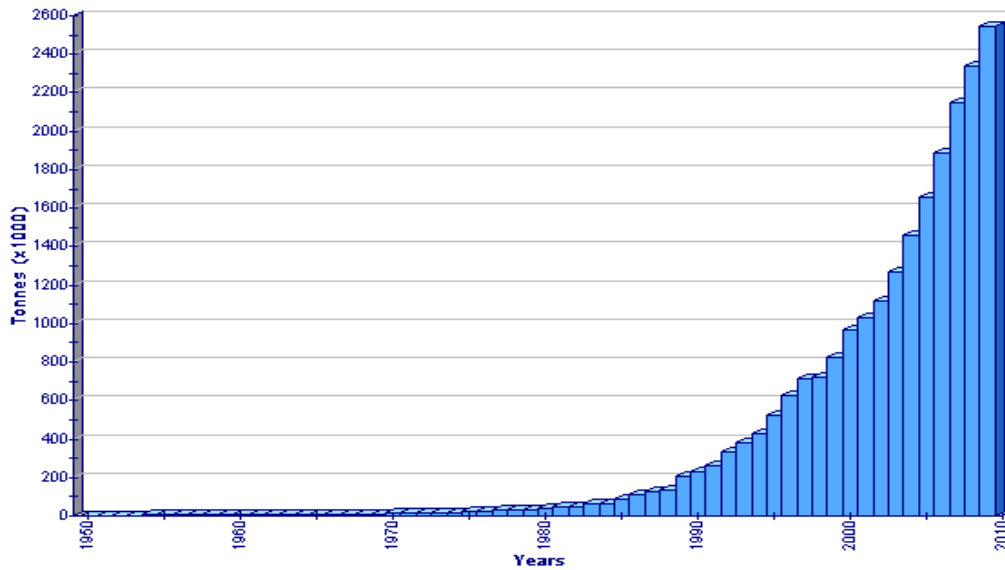


Figure 1 Global aquaculture production of Nile tilapia (Source: FAO, Fishery statistics, 2010).

Comparison of major farmed fishes

Species	Geography	Consumers	Fish meal	Systems	Freshwater or Marine
Salmon	Regional	Global	Moderate	Cages	Requires both
Carps	Global	Regional	Minimal	Ponds & cages	Freshwater only
Catfish	Global	Global	Minimal	Ponds & cages	Freshwater only
Sea bass, cobia, snappers	Global	Global	High	Cages, recirc systems	Marine only
Tunas	Regional	Global	High	Cages	Marine only
Tilapia	Global	Global	Minimal	Ponds, cages, raceways, recirc systems	Either

Figure 2. Global comparison of tilapia and other major farmed fishes (From Fitzsimmons, 2010).

On the way to production technological innovation of tilapia farming practice, the Genetically Improved Farmed Tilapia (GIFT) project has demonstrated that using selective breeding enhance the growth performance of Nile tilapia by 80% from the base population after five generation (World Fish Centre, 2004). Growth performance is further improved through selective breeding program (Eknath et al., 2007). The GIFT strain is widely available for farmer in East and South-East Asia. Presently, Genomar Supreme Tilapia (GST) has being developed from GIFT tilapia. GST is more superior than the original GIFT strain and has better growth, feed conversion ratio, fillet yield and disease resistance compared to wild tilapia (Genomar, 2009).

Despite, genetic and environmental factors (Charo-Karisa et al., 2006), quality feed i.e. low cost and high nutritional value also play vital role for maximum growth and farm benefit of tilapia. In intensive farming systems, feed is the major cost item, according to Tacon and De Silva (1997) feed makes 70% of total cost for world fin fish production and Bostock et al. (2010) also gave emphasis on external feed as a factor for future intensification of aquaculture farming along with water and energy. High quality animal protein, lipid and other essential nutrients are required for intensive aquaculture (Tacon et al., 2010). To reduce the feed cost, several efforts has been made to replace the expensive feed ingredients. Fish meal (FM) has been partially or completely replaced by plant proteins though some conflicting results were evident in a review study by El-Saidy and Tacon (1997). Recent study using 5% fish meal and soybean meal replacing with cotton seed meal showed that 75% soybean meal can be replaced by cotton seed meal without any effect on growth performance of fingerling tilapia (El-Saidy et al., 2011). In another study, Zhao et al. (2010) found that fish meal can be completely replaced by soy protein concentrate by increasing feeding frequency for Nile tilapia less than 2 g. And Monentcham et al. (2010) showed that fish meal can be replaced up to 50% by soybean and cotton seed meal mixer (1:1 ratio) in a study on fingerling of *Heterotis nilotica*. Fish meal can also be completely replaced by extruded and full-fat soybean meal supplemented with methionine and lysine for Nile tilapia (Goda et al., 2007).

Supplementation of essential amino acid in mixture diet of fish meal and plant protein improved growth performance of Nile tilapia (El-Dahhar and El-Shazly, 1993).

Therefore, plant proteins are being widely used with fish meal for tilapia farming with or without supplementation of essential amino acid. The plant protein has anti-nutritional factors and imbalanced amino acid profile that negatively affect fish growth performance (Francis et al., 2001). But these issues have already been well addressed by using different processing methods on the ingredients, e.g. extracted and toasted soybean meal is commercially available with reduced anti-nutritional factors.

The optimum protein requirement of Nile tilapia depends on size, age, and water temperature. Several studies has been estimated that protein requirement for juvenile tilapia varies from 32 to 50% and for larger tilapia 25 to 30% (Hafedh, 1999; Nguyen, et al., 2009; El-Saidy et al., 2005; Ali et al., 2008; Abdel-Tawwab et al., 2010; Gunasekra et al., 1996; NRC., 1993).

The optimum dietary lipid requirement for tilapia is 5 to 12% (Lim et al., 2011), and Han et al. (2010) found significantly better growth by increasing dietary lipid from 55 to 85 g per kg diet. According to Lim et al. (2011) tilapia require linoleic (n-6) series fatty acids (18:2n-6 or 20:4n-6) and it can enhance the growth better than the n-3 series (18:3n-3, 20:5n-3 or 22:6n-3).

Tilapia utilize starch efficiently from 22 to 46% dietary starch while 22% considered as optimum level for juvenile tilapia (Wang et al., 2005). So, the growth of tilapia can be enhanced by using optimum protein, lipid, carbohydrate and other nutrients also has similar type influence on growth performance of tilapia.

In addition, it has been evident from several studies that feeding rate and meal frequency can influence the production performance of tilapia. Study with polyculture farming of tilapia, common carp and silver carp showed that growth performance, body fat and gross energy gain increased as feeding rate (0 to 5% and to apparent satiation) increased (Abdelghany and Ahmad, 2002). Tambaqui showed better outcome using 10% feeding rate and 3 meals per day at growth phase (Silva et al., 2007). Research from pikeperch (6.4 g) give enhanced growth at 2% feeding rate and 3 meals/day (Wang et al., 2009). Yuan et al. (2010) found increased growth performance, protein and lipid contents with increasing feeding rate and Riche et al. (2004b) reported that growth efficiency of tilapia

increased if they allowed for four hours satiation feeding. Increased daily feeding rates from 30 to 60% of body weight for juvenile tilapia (12 mg body weight) gave significantly higher growth (Santiago et al., 1987). A study with red tilapia showed that best growth can be achieved near satiation feeding rate (Clark et al., 1990). According El-Saidy et al. (2005), tilapia with average weight 61.9 g showed cost effective and affordable feed strategy at 2% feeding rate. Similarly, Storebakken and Austreng (1987) found that Atlantic salmon showed increased growth by increasing the ration level from 0.5 to 1.0 of expected appetite level, but further increase in ration doesn't support for the growth.

Increased meal frequency provided better carbohydrate utilization for hybrid tilapia (Tung and Shiau, 1991). Photoperiod also influences the growth of tilapia and El-Saidy and Kawanna (2004) stimulated the growth of tilapia growth by using longer photoperiod.

Therefore, the present study was designed to investigate the effects of feeding rate on growth performance, body composition, nutrients and energy retention and apparent digestibility in Nile tilapia at fixed feeding frequency and fixed longer photoperiod. In the experiments, day length was kept long and feeding frequency high in order to eliminate these two as limiting factors for feed intake and growth.

Declining feeding rates were considered for juvenile tilapia. Relative feed intake decreases as the tilapia body weight increase and growth rate of smaller tilapia are higher than larger tilapia (Xie et al., 1997a).

Sub objectives:

- To find out how feeding rate affected growth performance, body composition, nutrients and energy retention and apparent digestibility of adult Nile tilapia (Experiment 1).
- To find out how feeding rate affected growth performance, body composition, nutrient and energy retention of juvenile Nile tilapia (Experiment 2).

- To find out how declining feeding rate affected growth performance, body composition, nutrient and energy retention of juvenile Nile tilapia (Experiment 3).
- To investigate effects of feeding rate on growth performance, body composition, nutrient and energy retention of Nile tilapia at early sexual maturation stage (Experiment 4).

CHAPTER TWO

2. Materials and methods

The research was carried out at the Fish Nutrition Laboratory, UMB, Ås Norway.

Nile tilapia is being raised at Fish Nutrition Laboratory which was generated from Genomar Supreme Tilapia (GST, Generation 16 of genetic selection). The GST is originated from genetically improved farmed tilapia (GIFT) project.

Four experiments were conducted in order to investigate effect of feeding rate on growth performance, body composition, nutrient and energy retention and apparent crude protein digestibility of Nile tilapia. Each tank contain 10 fish in Expt.1, 100 fish up to two weeks then after 50 fish in Expt.2, 100 fish in Expt.3 and 20 fish in Expt.4.

The 1st experiment was designed using 2.25% feeding rate (Tran-Duy et al., 2011) to define anticipated satiation level. The 2nd experiment was planned with anticipated appetite level at 8% and 6% (Fig. 4), respectively for 1st and 2nd fortnight, based on a pre-trial. The 3rd experiment was designed based on declining feeding rate (Table 4, Fig. 5) to compare report found for juvenile tilapia (1.1g) of Expt.2. Expt. 4 was designed by using feeding rate 1-5%. Two types of tanks (big tank, 210 L and small tank, 115 L water volume) with recycling water (Appendix 1) were used for all experiments. Detail of initial mean weight, type of tank used, start and end date of four experiments are given in Table 1. Randomized designs were used to start the all experiments.

Table 1: Initial mean weigh, type of tank uses, start and end date and total duration of four experiments.

Experiment	Initial mean weight	Type of tank	Start date	End date	Total duration(day)
1	257.9±0.3	Big	04.08.10	15.09.10	42
2	1.1±0.02	Small	08.10.10	05.11.10	27
3	1.1±0.02	Small	10.11.10	07.12.10	28
4	77.9±0.03	Small	27.01.11	09.02.11	13

2.1. Production of tilapia feed.

Tilapia feed were prepared considering nutritional value using facility at the Centre for feed Technology at UMB including a twin screw extruder.

2.1.1 Feed formulation:

Only one nutritionally balanced feed (Table 2 and 3) was prepared for all the four experiments. The feed was prepared from fish meal, soybean meal, sunflower meal, pea protein concentrate, corn gluten and wheat were mixed with appropriate amount soy oil, vitamin and mineral premix, mono calcium phosphate, yttrium oxide, lysine and methionine (Table 2).

Table 2: Formulation of experimental feed.

Content	Quantity g Kg ⁻¹
Dry matter (DM), g kg-1	908.7
<i>Ingredients composition</i>	
Fish meal ^a ,	70.0
Soybean meal ^b ,	196.4
Sunflower meal ^c ,	204.0
Pea protein concentrate ^d ,	50.0
Wheat ^e ,	300.0
Corn gluten ^f ,	80.0
Soy oil ^g ,	77.0
Mono calcium phosphate ^h	10.0
Vitamin and mineral premix ⁱ ,	5.0
Yttrium oxide ^j ,	0.10
L-lysine ^k	2.0
Dl-methionine ^l	5.5

^a NorsECO-LT, Norsildmel, Egersund, Norway.

^b Denosoy, extracted and toasted soybean meal, Denofa, Fredrikstad, Norway.

^c Sunflower meal, Extracted sunflower, Ukrain.

^d Pea protein concentrate, Aquamarine Nutrition, Stavanger, Norway

^e Wheat, Feleskjøpet, Norway

^f Corn gluten, Feleskjøpet, Norway

^g Denofa, Norway.

^h Mono calcium phosphate, Feleskjøpet, Norway

ⁱ Contents per kg: Vitamin A 2500.0 IU; Vitamin D₃ 2400.0 IU; Vitamin E 0.2 IU; Vitamin K₃ 40.0 mg; Thiamine 15.0 mg; Riboflavin 25.0 mg; d-Ca-Pantothenate 40.0 mg; Niacin 150.0 mg; Biotin 3.0 mg; Cyanocobalamine 20.0 µg; Folic acid 5.0 mg; Pyridoxine 15.0 mg; Vitamin C: 0.098 g (Stay-C 35, ascorbic acid phosphate, DSM Nutritional Products, Basel, Switzerland); Cu: 12.0 mg; Zn: 90.0 mg; Mn: 35.0 mg; I: 2.0 mg; Se: 0.2 mg; Cd ≤ 3.0 µg; Pb ≤ 28.0 µg; total Ca: 0.915 g; total K 1.38 g; total Na 0.001 g; total Cl 1.252 g; Trouw Nutrition, LA Putten, The Netherlands.

^j Metal Rare Earth Limited, Shenzhen, China.

^k L-lysine HCl, 99% feed grade, CJ Indonesia, Jakarta, Indonesia.

^l DL-methionine, 99% feed grade, Adisseo Brasil Nutricao Animal Ltda, Sao Paulo, Brazil.

2.1.2 Feed Analysis

Chemical compositions of the diet are given in Table 3. Dry matter content of diet were determined as weight loss after drying the samples at 103°C until constant weight (ISO, 1983). Crude proteins (Kjeldahl N×6.25) were determined Kjeltec auto 1035/1038 system (Tecator, Sweden). Solvent Extraction (ASE) method was used to determine crude fat of diet. Ash contents were determined by heating at 500°C in muffle furnace. Starch was analyzed as glucose after starch hydrolysis with a heat tolerant amylo-glucosidase in accordance with the procedure of (McCleary et al., 1994). The sample were burned at 500°C in muffle furnace then dissolved in 1M HCl and finally analyzed by spectrophotometer (Bourke and Yanagawa, 1993) to determine total phosphorus. Bomb calorimeter was used to calculate energy contents of diet.

Table 3: Chemical composition of experimental diet.

Chemical composition	Experimental Diet
Dry matter, g (kg) ⁻¹	908.7
Crude protein, g (kg DM) ⁻¹	341.63
Crude fat, g (kg DM) ⁻¹	66.55
Ash, g (kg DM) ⁻¹	47.00
Starch, g (kg DM) ⁻¹	25.10
Total Phosphorous g (kg DM) ⁻¹	4.98
Energy MJ Kg ⁻¹ DM	18.77

2.1.3 Diet preparation

Macro ingredients of formulated diet were weighed using large weighing scale mean while micro ingredients were weighed using Sartorius analytical balance. To produce slow sinking tilapia feed, all macro ingredients were milled in a Münch Hammer mill (HM 21.115, Wuppertal, Germany) and grinded to particle size of 0.5 mm using 1 mm screen. The milled ingredients and micro ingredients were mixed homogenously in a small Dinnisen twin shaft mixer (Pegasus Menger 400 1, Sevenum, Holland) for 2 minutes. Then it was transferred to a mini feeder of extruded barrel (Twin screw Bühler BCTB 62 extruder) to produce slow sinking diet. Into the barrel, the compounded mixer of raw ingredients precooked with addition of hot water, shearing, pressure and finally heat generated before exit through the die.

2.2 Technically what was done?

2.2.1 Screw configuration of extruder.

The feed mashes were being passed through extruder barrel and suitable screw configurations (Fig.3) to produce slow sinking feed for Nile tilapia. Screw configuration of extruder were- 100, 3x80, 60, L20, 80, 60, L20, 80, 60, L20, 80, 60, L20, 100, 80, 60, R20, 60, 40. Total length was 1240 mm (Fig. 3).

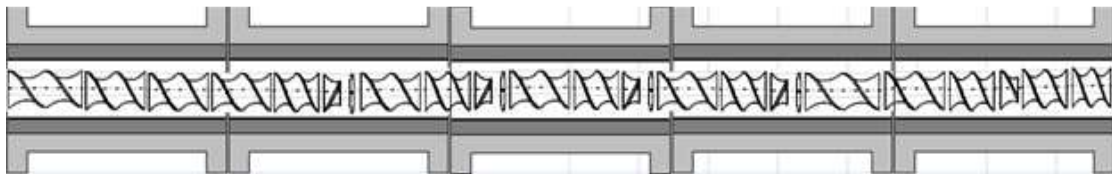


Figure 3. Screw configuration of extruded barrel for producing slow sinking feed for Nile tilapia.

2.2.2 Production of crumbled feed

The feed which had been produced through extruded barrel were hammered manually to reduce the pellet size, and then passed on sieving material of 2.8 mm, 2 mm, 1.5 mm and 1 mm pore size (Retsch, Germany) to get the desirable pellet size.

2.3 Feeding, water quality and standardization.

2.3.1 Feeding

In Expt. 1 feed were supplied six time in day by automatic feeder adjusted with for 3. hr and 40 minutes interval and 20 minutes feeding duration. And Expt. 2, 3 and 4 feed were supplied 22 times in day by automatic feeder and 55 minutes' interval and 5 minutes feeding duration. The feeding rate, frequency and photoperiod were adopted from (Riche et al., 2004a; Gjøen and Zimmermann., (Unpublished); Leal et al., 2010; Tran-Duy et al., 2011) to determine and evaluate for utilization of diets and growth condition of *O. niloticus* (257.9 g, 1.1g and 77.9 g at 26°C). Details of feeding treatment of Expt1, 2, 3 and 4 are given in Fig. 4,5,6 and Table 4.

All feeding treatment were subjected taking into account daily body weight increase based on expected FCR 1.1, 0.8, 0.8 and 0.9, respectively for experiment 1,2,3 and 4.

Table 4. Start and end feeding rate (% of biomass) with daily decrease of experiment 3

Treatment	Week 0-2		Week 3-4	
	Start feeding %	End feeding %	Start feeding%	End feeding %
Treatment 1	8	8	6	6
Treatment 2	10	8	8	6
Treatment 3	12	8	10	6
Treatment 4	14	8	12	6
Treatment 5	16	8	14	6

2.3.2 Water quality and standardization

According to experimental design, water quality like temperature should be $\geq 26^{\circ}\text{C}$, oxygen (O_2) $\geq 6 \text{ mg l}^{-1}$ and pH at 7. Temperature and oxygen were measured from installed OxyGurad Commander System in laboratory. In addition manual measurements were done by small OxyGuard instrument. Total ammonia nitrogen (TAN) and NO_2 (Nitrate) were measured at the end of experiment by LaMotte-Model NANR. Code 7418-02 and JBL Test NO_2 , respectively for TAN and NO_2 .

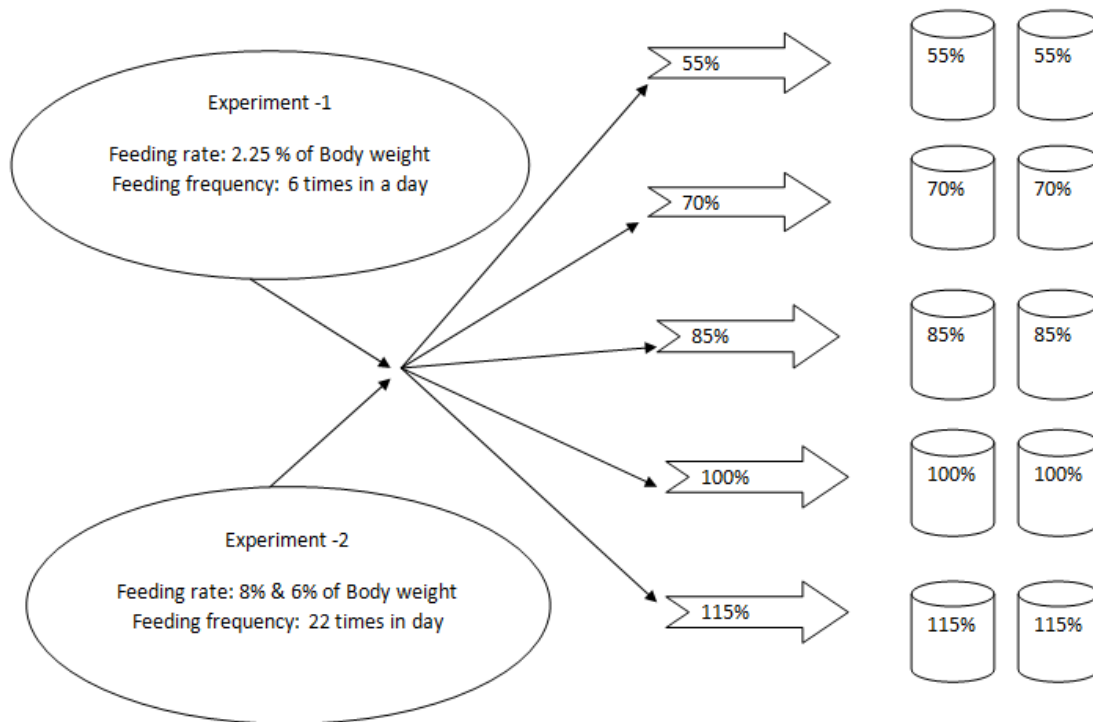


Figure 4. Layout of experiment 1 and 2; Expt.1 2.25% of body weight was considered as 100% feeding level while Expt. 2, 8% of body weight was considered 100% feeding level for first two week and 6% of body weight for last two weeks a satiation level.

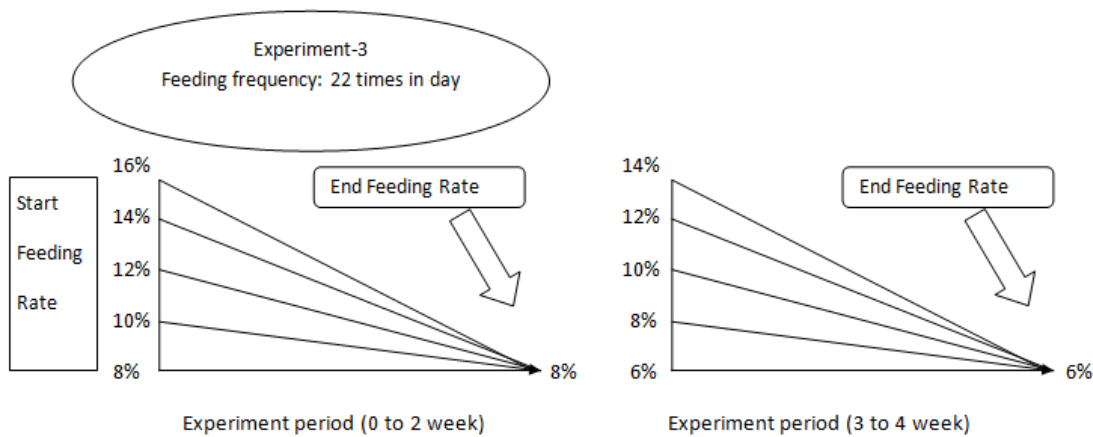


Figure 5. Layout of Experiment 3, Juvenile Nile tilapia fed with declining rate 8-16 to 8% and 6-14 to 6% designed to compare 8% and 6% feeding rate of experiment 2 during week 0-2 and 3-4 respectively a FCR at 0.8 g dry matter intake (g gain⁻¹)

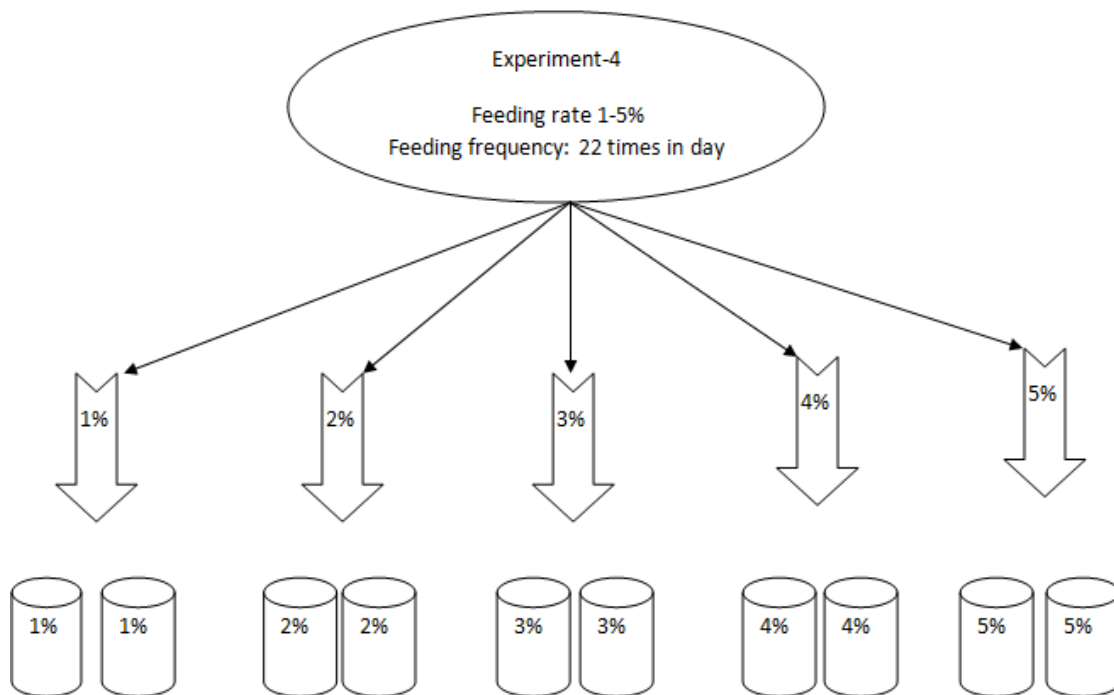


Figure 6. Layout of experiment 4 using feeding rate 1-5% body weight each treatment with duplicate treatment a FCR at 0.9 g dry matter intake (g gain^{-1}).

2.4 Weighing, sampling

During inception of the experiment, after weighing, some fishes (In Expt.1: 3; Expt.2 and 3: 10 and Expt. 4: 5 fish) were placed into the freezer, at -20°C . And at the end of the experiments, same numbers of fishes were placed at -20°C after weighing. Body weight of fishes was taken at 21 days interval (Expt.1); and fortnightly (Expt. 2, 3 and 4.). Before weighing, all fishes were anaesthetized by MS222 (0.2g l^{-1}). The faces were collected from distal part of intestine after opening of abdomen and also frozen for digestibility study.

2.5 Sample preparation

Meat grinder was used to grind the big fish while small grinder (A11 Basic Analytical mill, IKA, Wilmington, USA) was used for small fish. Grinded fishes and fecal content were subjected to freeze drier. To make the sample homogenous, it was treated with dry ice and then grinding was done. The feed samples were prepared by grinding the pellet

into mash by A11 basic Analytical mill. All the dry samples were kept at 4 °C until all analyses finished.

2.6 Analyses:

Dry matter, crude protein, fat, ash, total phosphorus and energy contents of fishes were measured by the method similar to section 2.1.2. In addition to these, Dumas method was used to measure fecal nitrogen content and inductively coupled plasma mass spectroscopy (ICP-MS) was used to measure yttrium oxide.

2.7 Calculation:

Weight Gain (WG %) were calculated by the following formula

$$WG\% = \frac{W_1}{W_0} \times 100$$

Where W_0 represent initial weight and W_1 final weight of the trial

Specific growth rate (SGR), were calculated according to following formula

$$G_w = \frac{\ln W_1 - \ln W_0}{T}$$

Where W_1 is final fish weight at the end experiment period, W_0 is the initial fish weight at the starting of the experiment and T is the time interval in days.

Feed conversion ratio (FCR) was calculated as:

$$FCR = F \times G^{-1}$$

Where F is the dry matter feed offered and G represents the weight gain.

Nitrogen and phosphorus retentions were calculated according to following formula:

$$\text{Nitrogen retention} = \frac{\text{Final nitrogen} - \text{Initial Nitrogen}}{\text{Nitrogen offered}} \times 100$$

$$\text{Phosphorus retention} = \frac{\text{Final Phosphorus} - \text{Initial phosphorus}}{\text{Phosphorus offered}} \times 100$$

Apparent digestibilities of crude protein were calculated according to following formula.

$$\text{Apparent digestibility of crude protein} = 100 \times \left(1 - \left(\frac{D_i}{F_i} \times \frac{F_n}{D_n}\right)\right)$$

Where D_i = Concentration of Y_2O_3 in diet

F_i = Concentration of Y_2O_3 in feces

D_n = Concentration of nutrient in diet

F_n = Concentration of nutrient in feces.

2.8 Statistical analyses:

All treatments were employed in duplicate: Significance level was $0.05 \leq P$. The results were analyzed by ANOVA in GLM (SAS, 1999) and second order polynomial regression in MS-Excel. Tukey's Studentized Range (HSD) test was used to rank the significant differences detected by ANOVA. The results are presented as least-square means and pooled standard errors of the means (S.E.M.).

CHAPTER THREE

3. Result

3.1 Experiment 1

In Expt. 1, Nile tilapias with an average start weight 257.9 ± 0.2 g were fed at feeding rate, ranging from 55 to 115% of an anticipated *ad libitum* level. This *ad libitum* level (feeding rate=100%) was defined by anticipated growth potential 2.25% body weight increase a day and a feed conversion ratio (FCR) of 1.1 g dietary dry matter per g gain.

3.1.1 Water quality

The temperature and pH (mean \pm S.E.M. total 42 days of measurement) were $25.9 \pm 0.5^\circ\text{C}$ (mean \pm S.E.M.) and 6.83 ± 0.0 , respectively during the whole experiment. Fig. 7 and 8 shows that oxygen, TAN (total ammonia nitrogen) and NO_2 (nitrite) were significantly affected by feeding treatment at the end of experiment when biomass in the tank was highest. The lowest and highest TAN content were found at 55% (0.15 ± 0.00 mg l^{-1}) and 115% (0.48 ± 0.03 mg l^{-1}) feeding level, respectively. On the other hand lowest and highest NO_2 was found 55% (0.25 ± 0.00 mg l^{-1}) and 115% (0.45 ± 0.05 mg l^{-1}), respectively.

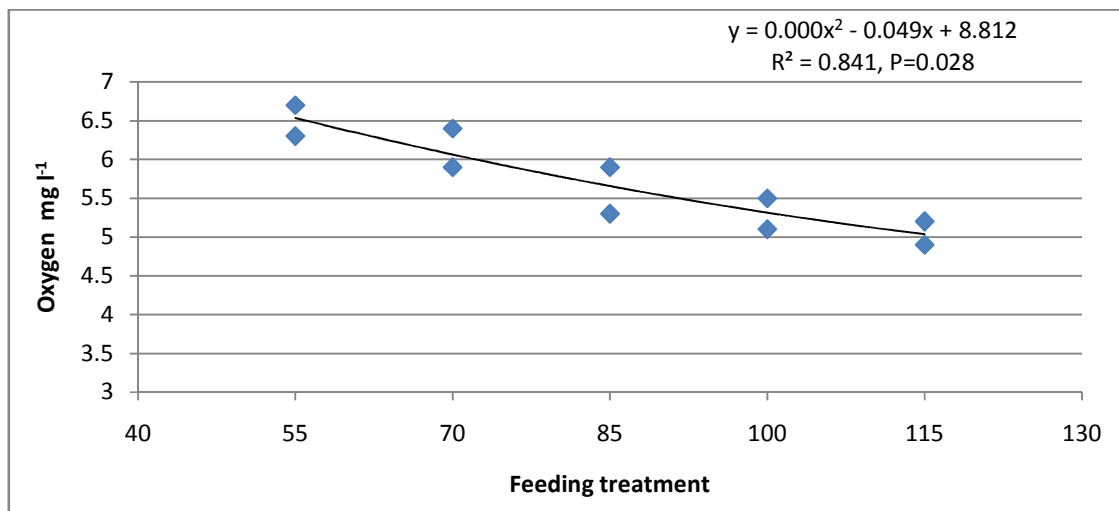


Figure 7. Oxygen, measurement in the rearing tank of Nile tilapia in experiment 1 at the end of experiment. 100% were defined by an expected daily weight gain at 2.25% and FCR at 1.1 g dry matter intake (g gain⁻¹).

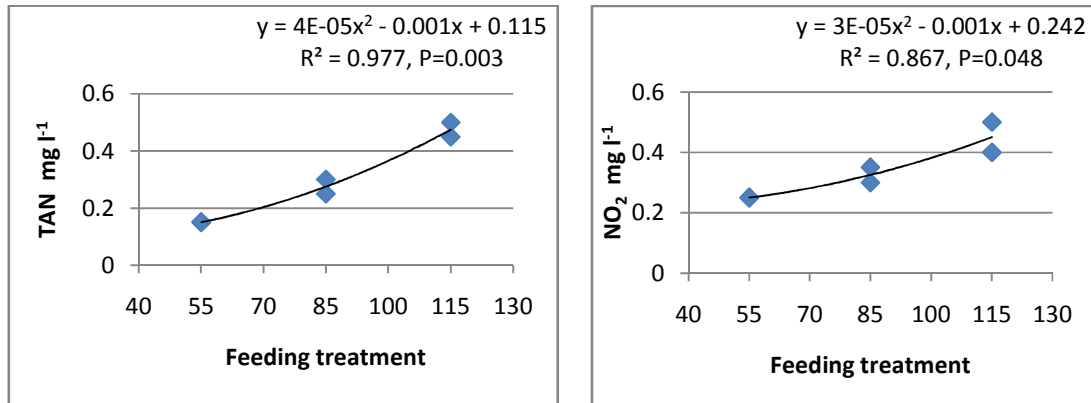


Figure 8. TAN and NO₂ measurement in the rearing tank of Nile tilapia in experiment 1 at the end of experiment. Measurement of TAN and NO₂ were only taken from feeding rate of 55, 85 and 115% where 100% were defined by an expected daily weight gain at 2.25% and FCR at 1.1 g dry matter intake (g gain⁻¹).

3.1.2 Growth parameters

Growth performance of the tilapia in Expt. 1 is shown in Table 5. Mean initial fish weights were not significantly different among feeding treatments. All of the fish survived during the experiment.

Weight gain and SGR during the first 3 weeks of feeding (Table 5, Fig. 9 and 13) tended (P=0.058 and 0.052) to be reduced for the 55% feeding rate, intermediate for 75%, while the tilapia seemed to utilize their growth potential for the feeding rates from 85 to 115%. The feed conversion ratios (FCR) varied from 1.0 g dry matter intake (g gain)⁻¹ for the tilapia on the most restricted feeding to 1.4 for the ones fed in excess (Fig. 11), but the difference was not significant.

During the last 3 weeks of feeding (Table 5), only FCR tended (P=0.073, Fig.11) to be more efficient (FCR=1.1) for the fish on the most restricted feeding regime, while the fish fed at the feeding rates of 85% and up used as much as 2.1-2.7 g DM (g gain)⁻¹. For the whole 6-weeks period only FCR=1.0 (Table 5) seemed likely to be proficient (P=0.055, Fig. 12) at lesser feeding regime for 55% whereas others FCR 1.2 to 2.0 were obtained at feeding rate 70 to 115% ranged though there were not significance difference among these.

Table 5. Growth performance of the Nile tilapia in Expt. 1, fed from 55 to 115 % a feeding rate planned to give an expected daily weight gain of 2.25% and a feed conversion ratio at 1.1 g dry matter intake (g gain)⁻¹ for the 100% feeding rate

Period	Feeding rate%	Initial weight	Final Weight	Survival (%)	Weight Gain (%)	FCR g dry matter intake(g gain ⁻¹)	SGR
Week 0-3	55	258.0	331.6	100	128.5	1.00	1.20
	70	257.8	346.9	100	134.6	1.08	1.41
	85	257.9	364.3	100	141.3	1.14	1.65
	100	257.6	367.7	100	142.7	1.37	1.69
	115	258.0	379.5	100	147.1	1.39	1.84
	Pooled SE	0.30	7.86		2.97	0.09	0.10
	P value	0.920	0.065		0.058	0.146	0.052
Week 4-6	55	331.6	423.7	100	127.8	1.02	1.17
	70	346.9	445.7	100	128.5	1.32	1.19
	85	364.3	444.2	100	121.9	2.14	0.94
	100	367.7	453.1	100	122.9	2.75	0.97
	115	379.5	472.5	100	124.5	2.67	1.04
	Pooled SE	7.86	18.99		3.18	0.36	0.12
	P value	0.065	0.666		0.584	0.073	0.588
Week 0-6	55	258.0	423.7	100	164.2	1.01	1.18
	70	257.8	445.7	100	172.9	1.20	1.30
	85	257.9	444.2	100	172.2	1.57	1.30
	100	257.6	453.1	100	175.9	1.92	1.33
	115	258.0	472.6	100	183.2	1.95	1.44
	Pooled SE	0.30	18.99		7.24	0.17	0.10
	P value	0.920	0.666		0.652	0.055	0.653

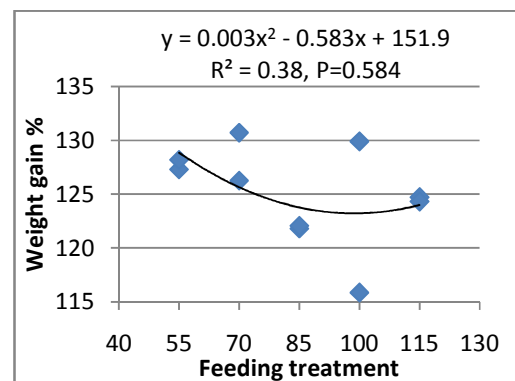
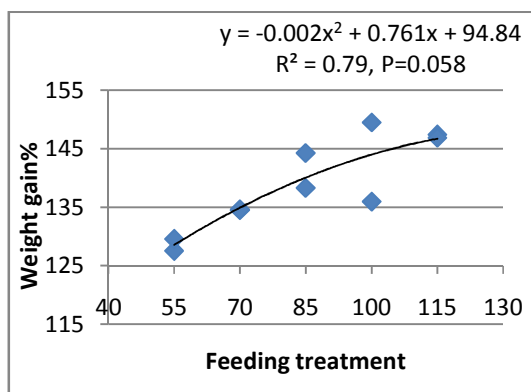


Figure 9. Weight gain in % during week 0-3(left side) and week 4-6 (right side). For definition of feeding treatment see Fig. 8.

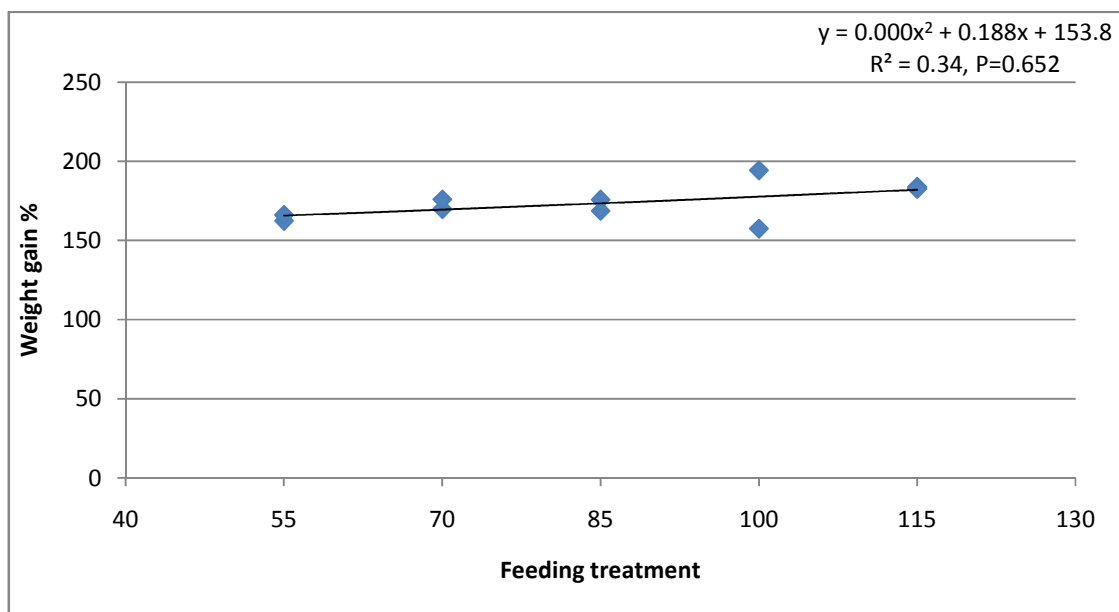


Figure 10. Weight gain in % during whole experiment period week 0-6. For definition of feeding treatment see Fig. 8.

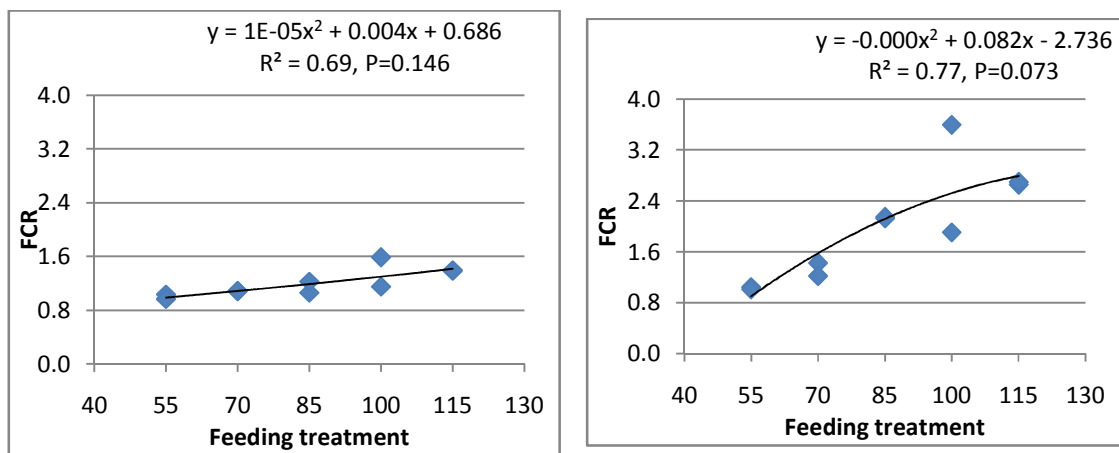


Figure 11. Feed conversion ratio (FCR) during week 0-3 (left side) and week 4-6 (right side). For definition of feeding treatment see Fig. 8.

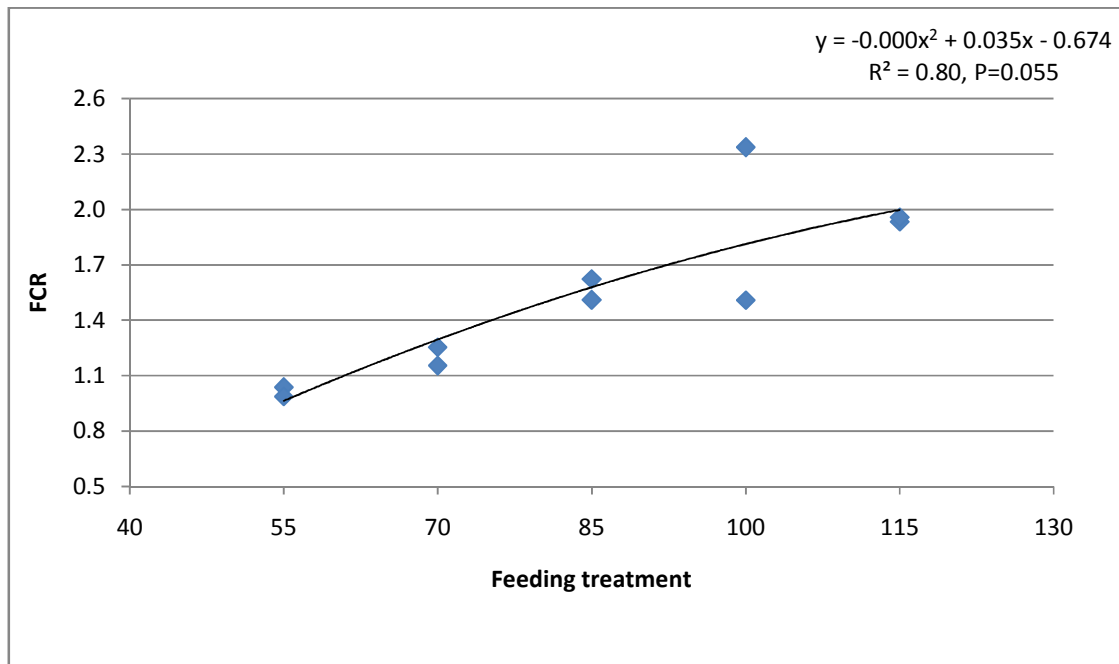


Figure 12. Feed conversion ratio during whole experiment period week 0-6. For definition of feeding treatment see Fig. 8.

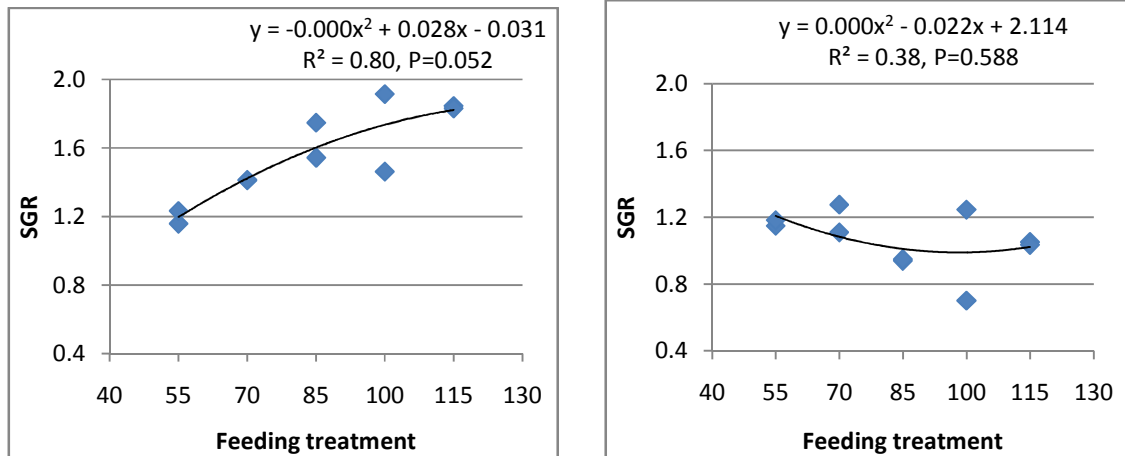


Figure 13 Specific growth rate (SGR) of experiment 1 during week 0-3 (left side) and week 4-6 (right side). For definition of feeding treatment see Fig. 8.

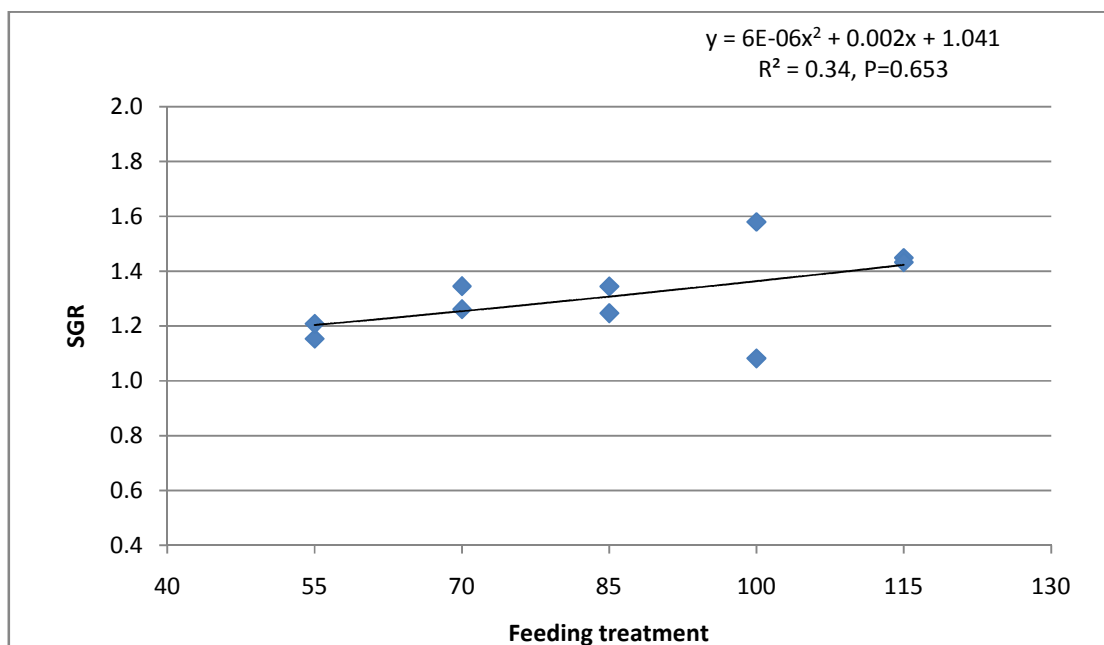


Figure 14. Specific growth rate (SGR) of experiment 1 during week 0-6. For definition of feeding treatment see Fig. 8.

3.1.3 Body composition

Crude fat tended to incline ($P=0.066$) to be abridged at 55% feeding rate (Table 6), intermediate 70%. Energy contents were affected by feeding treatment (table 6) and

Table 6. Body composition of the Nile tilapia in Expt. 1.

Experiment-1					
Fish	Dry matter (%)	Crude Protein (%)	Crude Fat (%)	Ash (%)	Energy (MJ kg ⁻¹)
Initial Body Composition	30.5	16.9	8.8	4.71	7.06 ^b
Final Body Composition					
55%	34.04	17.2	11.96	3.69	8.73 ^{ab}
70%	35.10	16.24	12.57	3.66	8.92 ^{ab}
85%	35.08	16.61	12.98	3.62	9.04 ^{ab}
100%	34.82	16.06	13.96	3.59	9.44 ^a
115%	34.35	16.76	15.16	3.51	9.8 ^a
Pooled SE	1.23	0.36	0.94	0.29	0.41
P value	0.186	0.389	0.066	0.150	0.036

found considerably higher than initial energy contents ($P=0.036$) where 100-115% feeding rate were significantly higher, intermediate at 55-85% than initial energy contents. There were no significant difference between 100 and 115% feeding rates and among 55-85% feeding rate ($P>0.05$).

3.1.4 Nutrients and energy retentions

Nitrogen retentions were predisposed to decline (Table 7, Fig. 15.) as the feeding level progressively increased ($P=0.001$) and were significantly higher at 55%, intermediate 70% than 85-115% feeding rate ($P<0.05$). There were non-significant difference between 100 and 115% feeding rate ($P>0.05$). Energy retention is tended ($P=0.065$) to be elevated at restricted feeding regime (Table 7) at 55% feeding rate (52.2%) where others were intermediate to higher feeding regime (70-115%) ranged from 44.5 to 31.1%.

Table 7. Nitrogen, phosphorus and energy retentions of Nile tilapia Expt. 1.

Feeding treatment	Nitrogen retention (%)	Phosphorus retention (%)	Energy retention (%)
55%	42.6 ^a	89.1	52.2
70%	35.3 ^{ab}	81.4	44.5
85%	27.3 ^{bc}	56.7	35.1
100%	23.1 ^c	37.9	32.3
115%	19.2 ^c	36.2	31.1
Pooled SE	1.78	16.40	4.09
P value	0.001	0.299	0.065

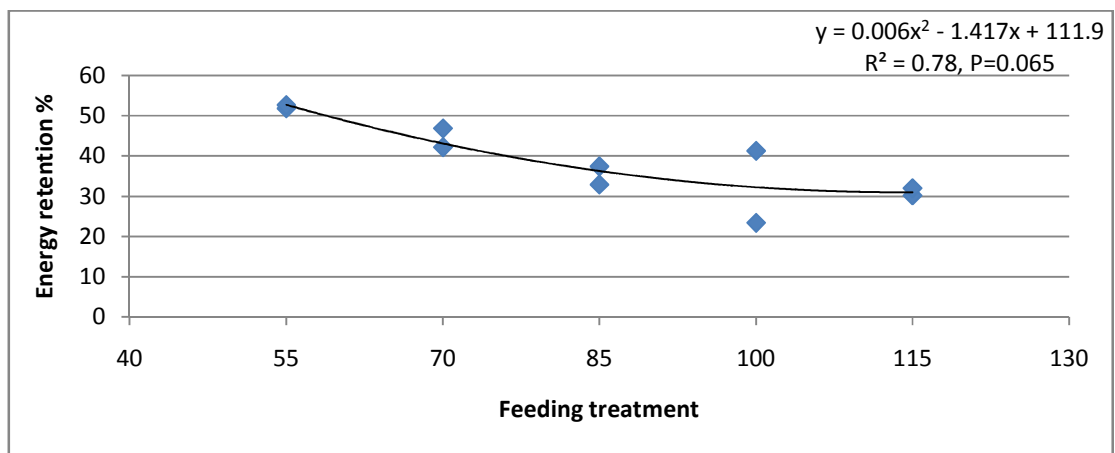
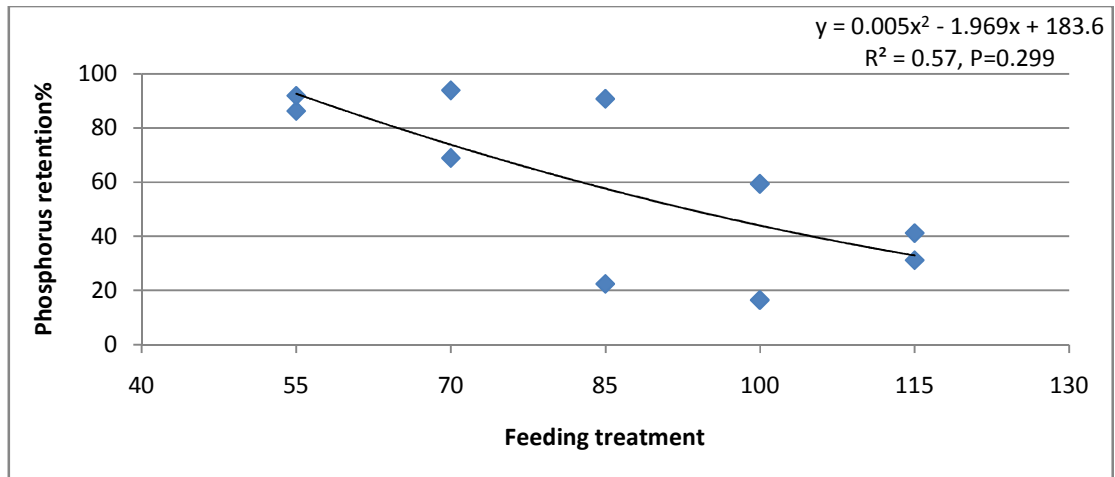
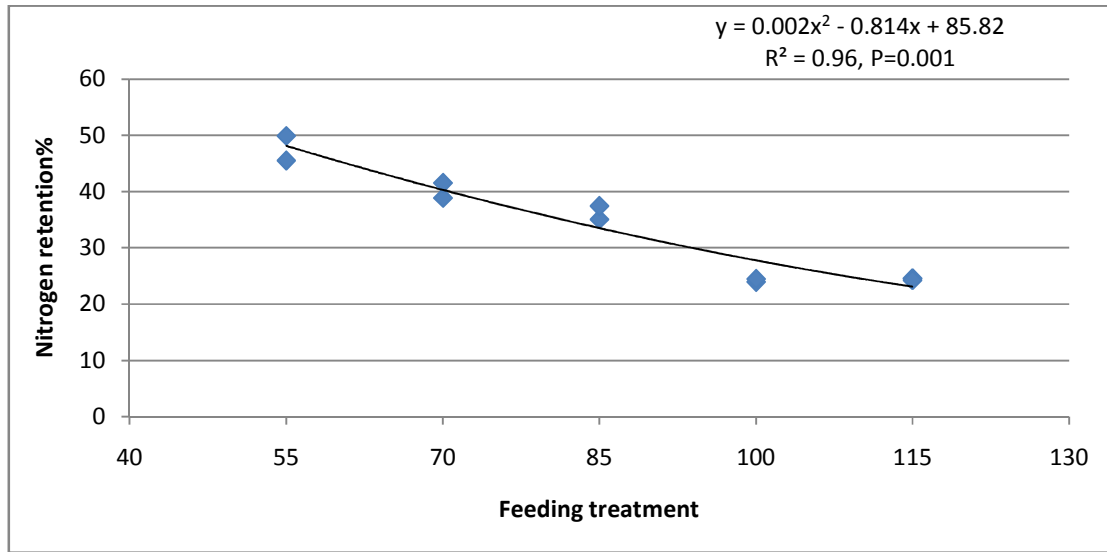


Figure 15. Nitrogen, phosphorus and energy retention of experiment 1. For definition of feeding treatment see Fig. 8.

3.1.5 Apparent protein digestibility

Apparent crude protein digestibility of Expt. 1 fed with 55-115%, were significantly different ($P=0.037$, Fig.16) of which highest crude protein digestibility were found at 100% (92.22 ± 2.71) feeding rate, while intermediate at 70, 85 and 115% feeding rate.

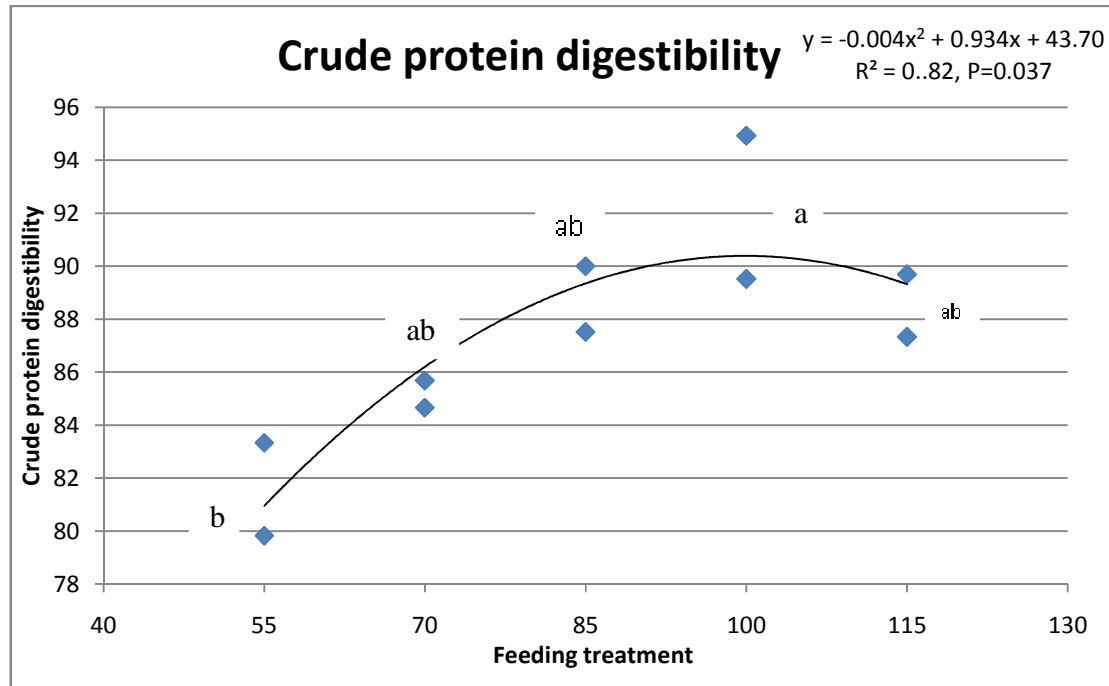


Figure 16. Crude protein digestibility of Exp.1. For definition of feeding treatment see Fig. 8.

3.1.6 Effect of feeding rate on sexual maturation and sexual maturation on growth performance.

Feeding rate and frequency have larger impact on sexual maturities which were observed through uneaten feed and available eggs in tanks (Observed during flush the tank water). Even there were some hatched out fry observed at the end of 21 days. There were no uneaten feed at 55% feeding level and the available uneaten feed and egg are given in Fig. 17. It shows that frequency of tracing uneaten feed and eggs were positively related with feeding rate. Fig. 9 and 13 showed that weight gain% and SGR were progressively decreased during last 21 days.

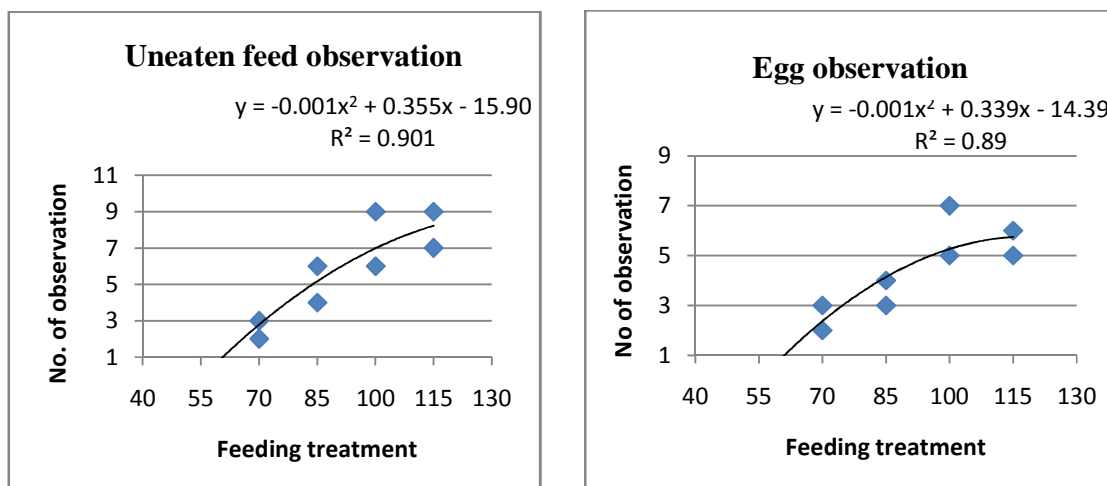


Figure 17. Observation of uneaten feed and egg during whole period of Expt. 1. For detail of experiment see Fig. 8.

3.2 Experiment 2

In Expt. 2, Nile tilapias with an average start weight 1.1 ± 0.0 were fed at feeding rate, ranging from 55 to 115% of an anticipated *ad libitum* level. This *ad libitum* level (feeding rate=100%) was defined by anticipated growth potential 8% body weight increase a day for 1st fortnight and 6% for 2nd fortnight and a feed conversion ratio (FCR) of 0.8 g dietary dry matter per g gain.

3.2.1 Water quality parameters

The temperature and pH of juvenile Nile tilapia experiment 2 were found as $25.5 \pm 0.5^\circ\text{C}$ and 6.83 ± 0.0 , respectively (Total 28 measurements were taken). The oxygen, TAN and NO_2 content were significantly ($P=0.009, 0.011, 0.014$, Fig. 18) affected by feeding regime. The highest O_2 was found at lowest feeding rate (55%, $6.85 \pm 0.05 \text{ mg l}^{-1}$) and lowest were found at highest feeding rate (115%, $5.15 \pm 0.05 \text{ mg l}^{-1}$). Meanwhile TAN and NO_2 increased as feeding rate progressed, lowest and highest TAN were obtained at 55% ($0.18 \pm 0.03 \text{ mg l}^{-1}$) and 115% ($0.55 \pm 0.05 \text{ mg l}^{-1}$) and NO_2 content were at 55% ($0.25 \pm 0.05 \text{ mg l}^{-1}$) and 115% ($0.68 \pm 0.02 \text{ mg l}^{-1}$), respectively.

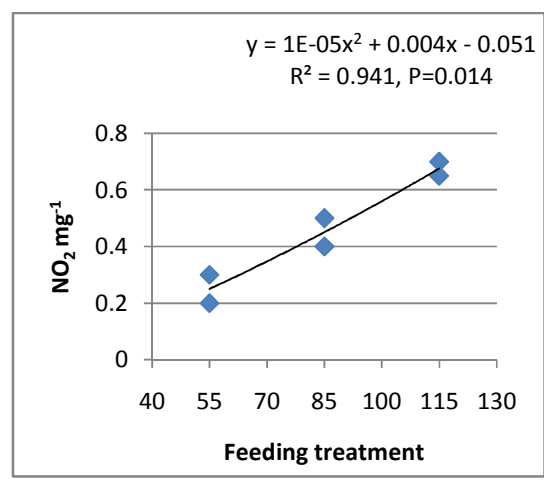
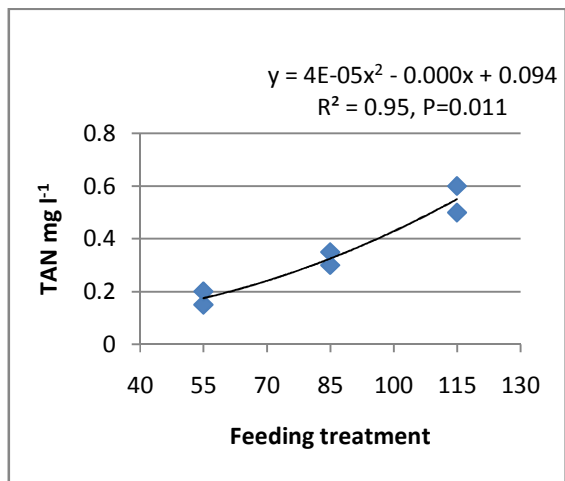
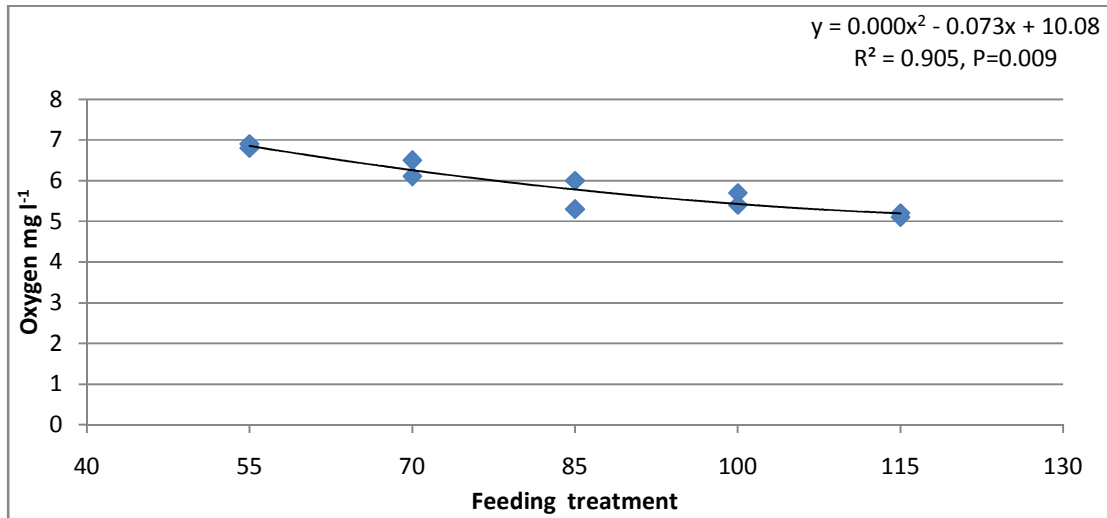


Figure 18. Oxygen, TAN and NO₂ measurement in the rearing tank of juvenile Nile tilapia in Expt. 1. Measurement of TAN and NO₂ were only taken from feeding rate of 55, 85 and 115% where 100% were defined by an expected daily weight gain at 8% and 6% respectively week 0-2 and week 3-4 and FCR at 0.8 g dry matter intake (g gain⁻¹).

3.2.2 Growth parameters of juvenile Nile tilapia

Growth performances of Juvenile Nile tilapia experiment 2 are given in Table 8. Initial weights were not significantly different (P=0.753). End of 1st fortnight fish weight were significantly different (P=0.014) due to feeding regime (55-115%) where limited feeding rate (55%) were significantly lower, intermediate at 70-85% and higher weight were obtained at uppermost(100-115%) feeding regime (Table 8). Survival rate were not significantly differed though there were some mortality in lowest and excess feeding rate

only during 1st two week. At the end of last two week of the experiment 2, fish weights were significantly higher at 85-115% and lower at limited (55-70%) feeding rate.

Weight gain (%), FCR and SGR were significantly affected by experimental feeding regime ($P= 0.006, 0.018$ and 0.008 respectively (Table 8). Weight gain (%) and SGR were notably higher as expected at 100% while intermediate at 85 and 115% feeding rate during 1st two week (Table 8, Fig. 19 and 23) while restricted feeding regime (55-70%) were significantly lower. Fig.21 and 22 show that FCR augmented to progressively increase as feeding rate gradually increased and were significantly lower at restricted feeding regime (55-85%) ranged from 0.65 to 0.72 g dry matter intake (g gain)⁻¹ and significantly higher at 115%, intermediate at 100% feeding rate (Table 8) during 1st two week.

At the end of last two week period, fish weight were significantly differed ($P\leq 0.001$, Table 8.) and no mortality were encountered where highest fish weight were gained at 85-115% feeding rate and trifling gain at 55-70% feeding rate. Consequently highest weight gain% (255.2%) and SGR (6.69) were obtained at 85% feeding rate and significantly higher than all other feeding rate while secondary at 70, 100 and 115% feeding rate and considerably lowest at 55% ($P=0.001$, Table 8). FCR are mostly affected by all feeding regime where all feeding rate were significantly differed with each other ($P\leq 0.001$, Table 8) treatment at 2nd two week period in which FCR (0.85) were obtained at 85% feeding rate that are significantly lower than 100-115%, and intermediate at 70% and considerable lowest at 55% feeding rate (Figure 21 and 22).

Taking into account of whole experiment 2 period, weight gain% (Fig. 20) and SGR (Fig. 24) particularly higher at upper (85-115%) and deprived at lesser (55-70%) feeding regime while FCR were very much lower (0.75 to 0.81) at 70-85% compare to intemperance supply regime (100-115%) that are assortment from 1.09 to 1.30 moreover lowest FCR (0.66) were brought into being at 55% feeding rate ($P\leq 0.001$, Table 8)

Table 8. Growth performance of the juvenile Nile tilapia in Experiment 2, fed from 55 to 115 % a feeding rate planned to give an expected daily weight gain of 8% and 6% during week 0-2 and week 3-4 respectively a FCR at 0.8 g dry matter intake (g gain)⁻¹ for the 100% feeding rate

Period	Feeding rate%	Initial weight(g)	Final Weight(g)	Survival (%)	Weight Gain (%)	FCR g dry matter intake(g gain ⁻¹)	SGR
Week 0-2	55	1.14	2.47 ^b	99	218.0 ^c	0.65 ^b	6.45 ^c
	70	1.11	2.72 ^{ab}	100	246.4 ^{bc}	0.73 ^b	7.14 ^{bc}
	85	1.11	3.31 ^{ab}	100	298.3 ^{ab}	0.72 ^b	8.53 ^{ab}
	100	1.11	3.61 ^a	99	324.9 ^a	0.82 ^{ab}	9.16 ^a
	115	1.14	3.50 ^a	99	307.9 ^{ab}	1.14 ^a	8.90 ^{ab}
	Pooled S.E.M.	0.02	0.19		17.19	0.09	0.43
	P value	0.753	0.014		0.006	0.018	0.008
Week 3-4	55	2.59 ^c	5.42 ^b	100	209.4 ^c	0.67 ^d	5.28 ^c
	70	2.75 ^c	6.25 ^b	100	227.9 ^b	0.75 ^{cd}	5.88 ^b
	85	3.25 ^b	8.30 ^a	100	255.2 ^a	0.85 ^c	6.69 ^a
	100	3.59 ^{ab}	8.21 ^a	100	228.8 ^b	1.23 ^b	5.91 ^b
	115	3.79 ^a	8.65 ^a	100	228.3 ^b	1.39 ^a	5.90 ^b
	Pooled S.E.M.	0.09	0.43	0.00	11.43	0.07	0.35
	P value	≤0.001	≤0.001		≤0.001	≤0.001	≤0.001
Week 0-4	55	1.14	5.42 ^b	99	478.2 ^b	0.66 ^d	5.59 ^c
	70	1.11	6.25 ^b	100	566.2 ^b	0.75 ^c	6.19 ^b
	85	1.11	8.30 ^a	100	749.5 ^a	0.81 ^c	7.19 ^a
	100	1.11	8.21 ^a	99	738.8 ^a	1.09 ^b	7.14 ^a
	115	1.14	8.65 ^a	99	761.2 ^a	1.30 ^a	7.25 ^a
	Pooled S.E.M.	0.02	0.43		42.21	0.06	0.24
	P value	0.753	≤0.001		≤0.001	≤0.001	≤0.001

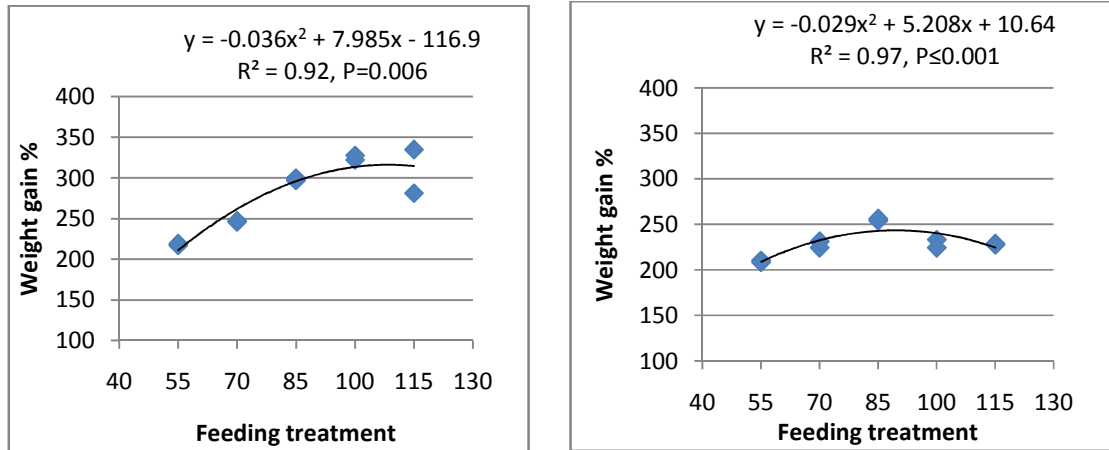


Figure 19. Weight gain % of experiment 2 during week 0-2 (left side) and week 3-4(right side). For definition of feeding treatment see Fig. 18

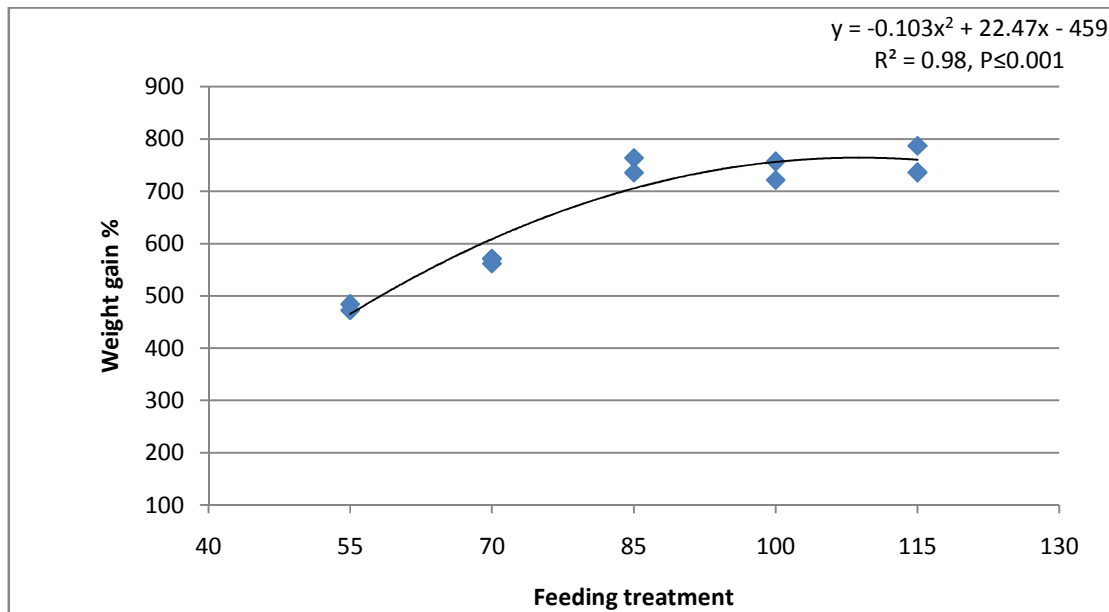


Figure 20. Weight gain % of Expt. 2 during week 0-4. For definition of feeding treatment see Fig. 18

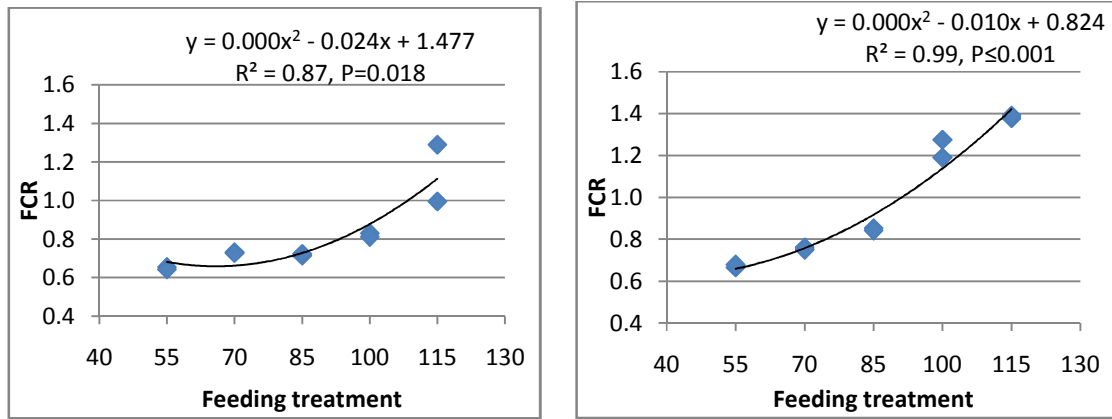


Figure 21. Feed conversion ratio (FCR) of experiment 2 during week 0-2 (left side) and week 3-4 (right side). For definition of feeding treatment see Fig. 18

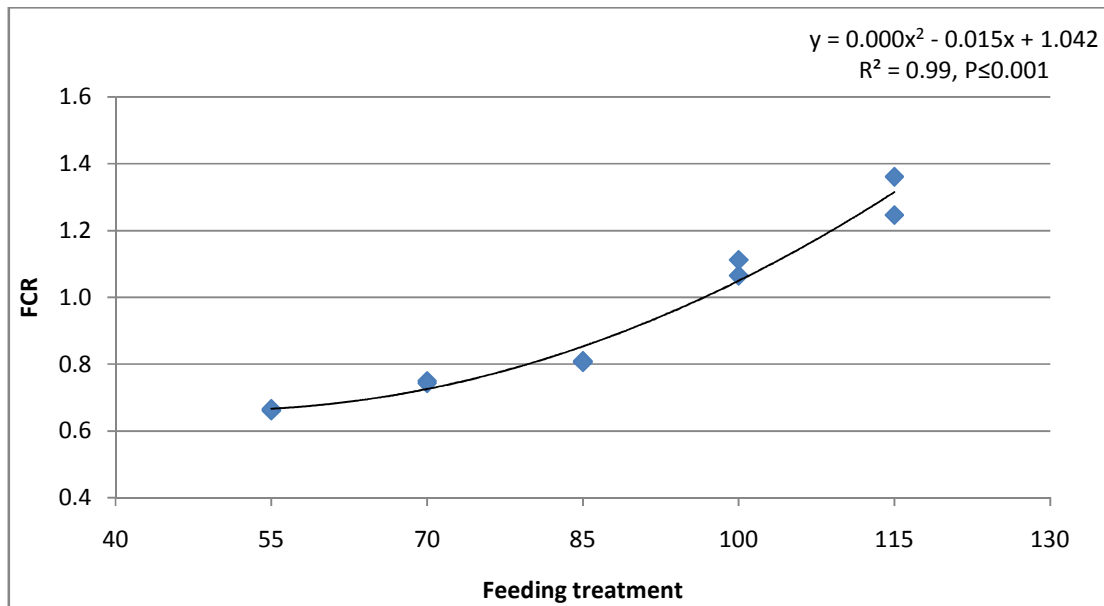


Figure 22. Feed conversion ratio (FCR) of experiment 2 during week 0-4). For definition of feeding treatment see Fig. 18.

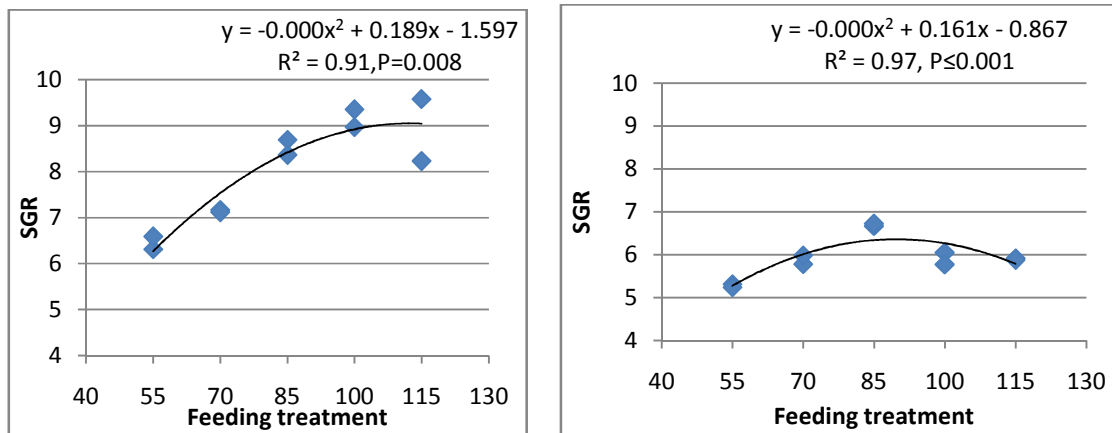


Figure 23. Specific growth rate (SGR) of experiment 2 during week 0-2 (left side) and week 3-4 (right side). For definition of feeding treatment see Fig. 18

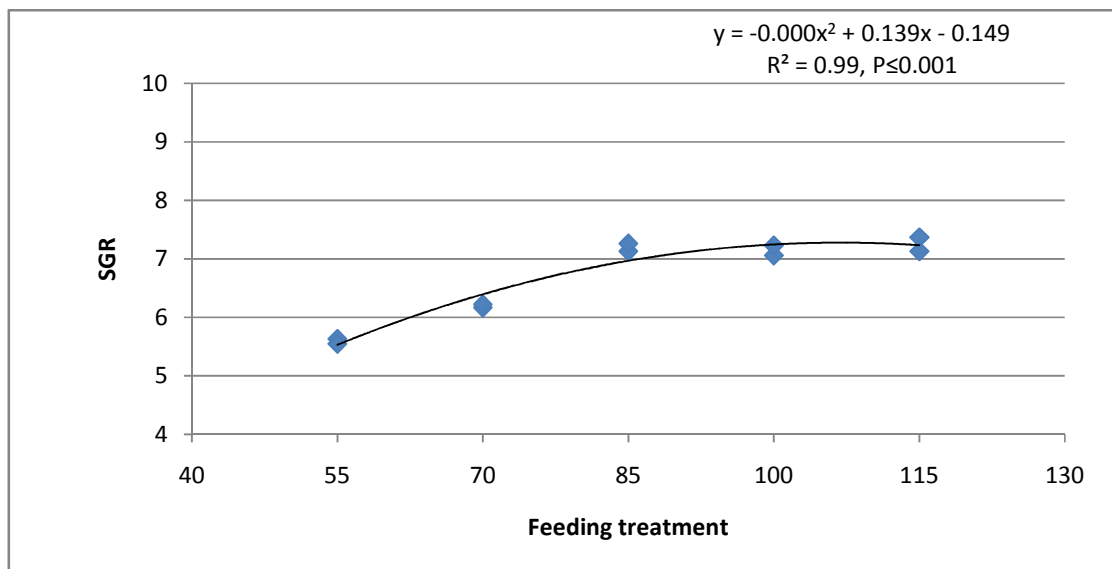


Figure 24. Specific growth rate (SGR) of experiment 2 during week 0-4. For definition of feeding treatment see Fig. 18.

3.2.3 Body composition of juvenile Nile tilapia.

The chemical composition of juvenile Nile tilapia belonging to Expt. 2 significantly differed on dry matter, crude protein and ash than initial body composition (Table 9). Crude fat were significantly higher at feeding regime (85-115%) from initial fat contents and intermediate at 70% feeding rate. Dry matter contents were significantly higher at 70, 100 and 115% feeding rate and intermediate at 55 and 85% feeding rate (Table 9). Ash

contents were considerably higher at 55-85, 115% and intermediate at 100% feeding rate. Experimental feeding regime did not significantly differed on energy content especially at 70, 85 and 115% feeding rate (Table 9) but 100% feeding rate were significantly higher than initial body composition of energy contents.

Table 9. Body composition of the juvenile Nile tilapia in Expt. 2, fed from 55 to 115 % a feeding rate planned to give an expected daily weight gain of 8% and 6% during week 0-2 and 3-4 respectively.

Fish		Dry matter (%)	Crude Protein (%)	Crude Fat (%)	Ash (%)	Energy (MJkg ⁻¹)
Initial Body Composition		25.96 ^b	12.30 ^c	9.69 ^b	1.82 ^b	7.09 ^{ab}
Final Body Composition						
	55%	28.04 ^{ab}	14.66 ^a	9.68 ^b	2.33 ^a	6.89 ^b
	70%	30.11 ^a	14.04 ^{ab}	11.37 ^{ab}	2.41 ^a	7.75 ^{ab}
	85%	28.98 ^{ab}	13.39 ^{bc}	12.32 ^a	2.26 ^a	7.83 ^{ab}
	100%	29.86 ^a	12.26 ^c	12.68 ^a	2.02 ^{ab}	8.43 ^a
	115%	30.74 ^a	13.62 ^{ab}	12.89 ^a	2.22 ^a	8.29 ^{ab}
Pooled SE		0.93	0.65	0.38	0.15	0.25
P value		0.018	0.001	0.002	0.007	0.029

3.2.4 Nutrient and energy retentions

Nitrogen, phosphorus and energy retentions were significantly differed by given feeding regime ($P \leq 0.000$, 0.001 and 0.001 respectively, Table 10.). According to Fig. 25, these are like to be decreased as feeding rate is raised. Nitrogen retention (%) were significantly differed at 55-85% feeding regime and 55% rate were found significantly higher than 70-115% feeding regime while there were significantly lower and not differed excess feeding (100-115%). Total phosphorus and energy retention were significantly higher at 55-85% given feeding treatment while there were significantly lower at excess feeding regime (100-115%).

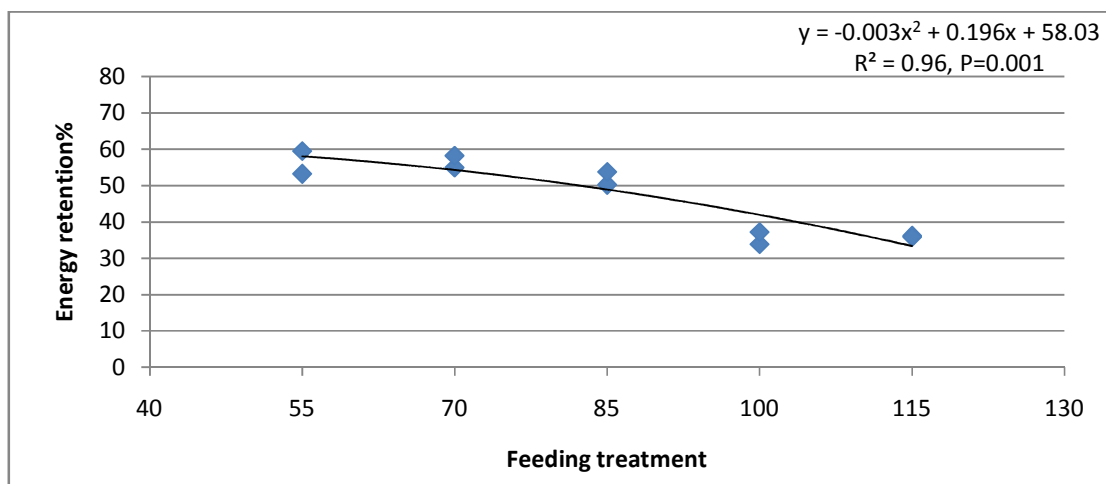
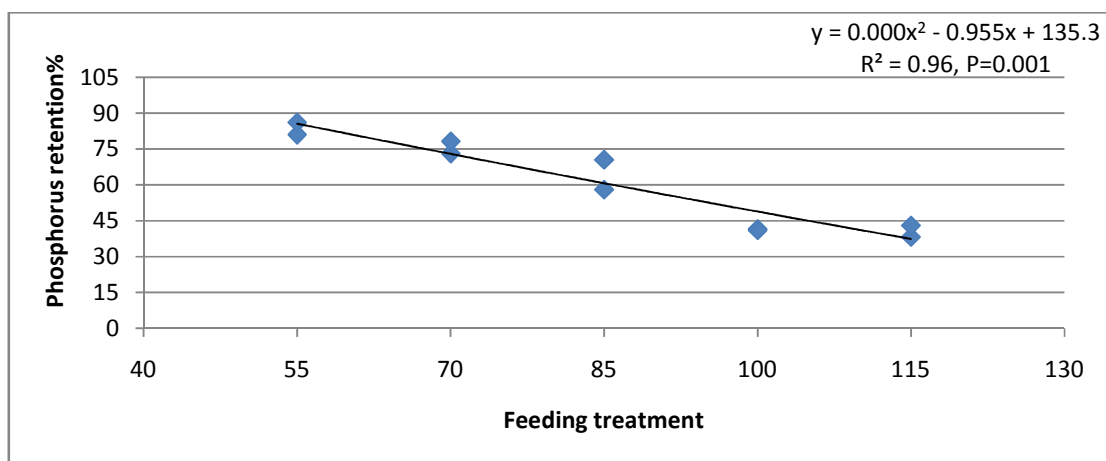
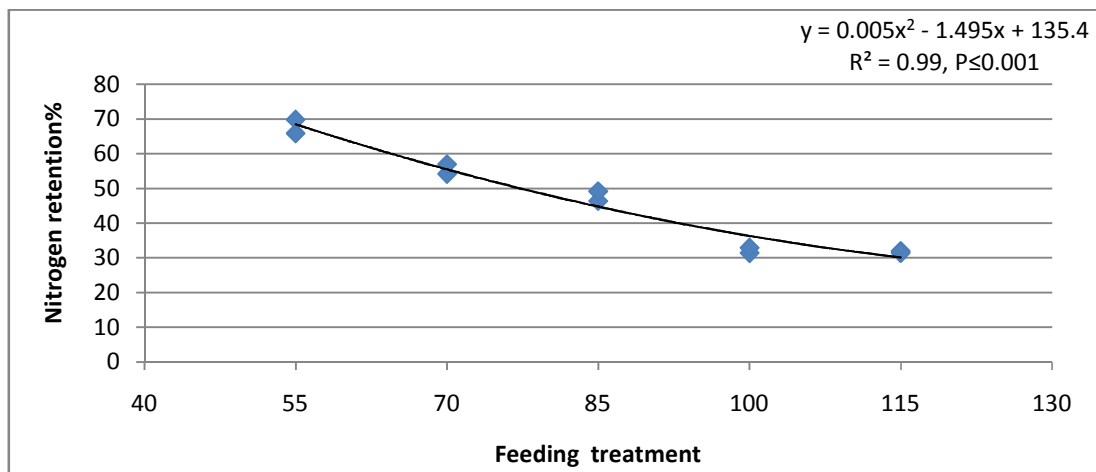


Figure 25. Nitrogen, phosphorus and energy retentions of Expt. 2. For definition of feeding treatment see Fig. 18.

Table 10. Nitrogen, phosphorus and energy retention of juvenile Nile tilapia Expt. 2 at 55-115% feeding rate planned to give 8% and 6% body weight gain during 1st and last two week respectively.

Feeding treatment	Nitrogen retention (%)	Phosphorus retention (%)	Energy retention (%)
55%	67.9 ^a	83.6 ^a	56.4 ^a
70%	55.6 ^b	75.7 ^a	56.7 ^a
85%	47.7 ^c	64.3 ^a	52.0 ^a
100%	32.2 ^d	41.3 ^b	35.6 ^b
115%	31.7 ^d	40.6 ^b	36.1 ^b
Pooled SE	2.76	4.54	3.58
P value	≤0.001	0.001	0.001

3.2.5 Uneaten feed

There were uneaten feed at one tank of 115% feeding rate last three days at the end of 1st two week and looked unusual compare to other tank eventually fishes were subjected to kill. Fish were divided from other 115% feeding rate tank. Experiment was continued as 50 fish per tank for rest of the period.

3.3 Experiment 3

In Expt. 3, Nile tilapias with an average start weight 1.1 ± 0.0 were fed at two declining feeding rate, starting 16-8% and end to 8% for 1st two week and starting 14-6% and end 6% for week 3-4 period a feed FCR of 0.8 dietary dry matter per g gain. This level was defined in order to check growth performance at 8% and 6% body weight increase a day for Expt. 2. Five treatments were taken to run the experiment and explanation of feeding treatment given in Table 4 and Fig. 5.

3.3.1 Water quality parameters

The temperature and pH were found as; $25.5 \pm 0.5^{\circ}\text{C}$ and 6.82 ± 0.0 respectively during whole experiment period (Total 28 days observation). Oxygen, TAN and NO_2 were significantly influence by feeding treatment ($P=0.004$, 0.013 and 0.004 respectively, Fig. 26). Fig. 26 shows that available of oxygen in the feeding tank were decreased as feeding

rate increased and lowest oxygen concentration were found excess feeding rate 115% (5.1 ± 0.3 mg/l) at the end of experiment. Lowest and highest TAN content were found at restricted 55% (0.28 ± 0.0 mg/l) and highest 115% (0.6 ± 0.0 mg/l) feeding rate respectively. At the same time lowest and highest NO_2 contents were found at treatment - 1 (0.1mg/l) and treatment-5 (0.4 mg/l) respectively

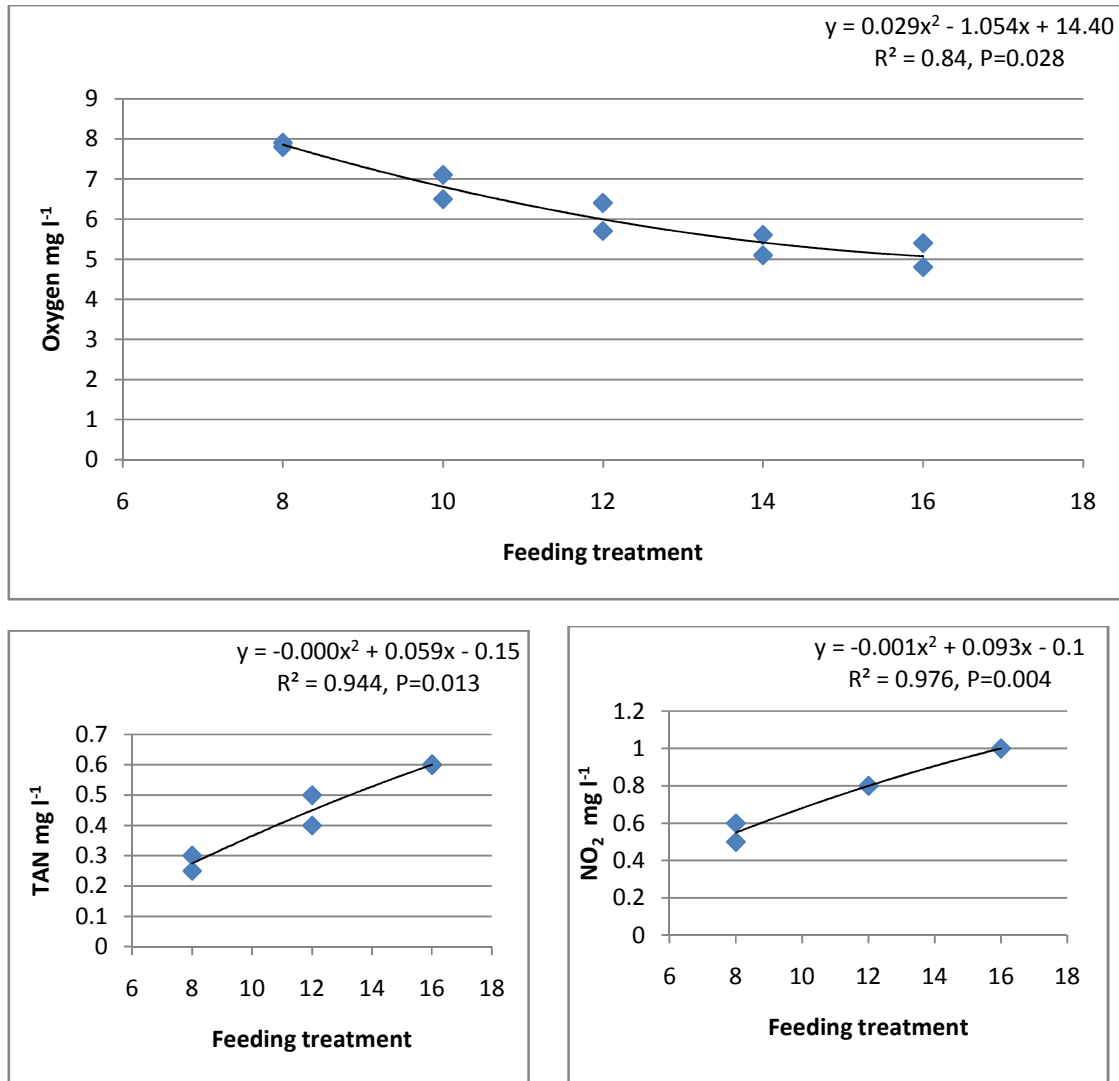


Figure 26. TAN and NO_2 concentration of different feeding treatment at the end of the 4-week feeding period in Expt. 3. Measurement of TAN and NO_2 were only taken from treatment 1, 3 and 5 at FCR of 0.8 dietary dry matters per g gain. This level was defined in order to check growth performance at 8% and 6% body weight increase a day for Expt.2. Feeding rates 8, ..., 16 in the figure defines feeding rates during first 2 weeks.

3.3.2 Growth parameter

Initial weight of Expt. 3 did not differ significantly ($P=0.48$, Table 11). Fish weight at the end of 1st two week significantly differed ($P=0.004$). All fishes were survived except treatment 3 (Survival 99%) after 1st two week period. Weight gain%, FCR and SGR significantly differed by the feeding treatment ($P\leq 0.001$, Table 11). Highest and intermediate weight gain% and SGR were obtained at feeding treatment 4 and 3, respectively during 1st two week (Fig. 27 and 31).

Table 11. Growth performance of the juvenile Nile tilapia in Experiment 3, fed with declining rate 8 -16 to 8 % and 6-14 to 6% designed to check 8 and 6% weight gain of experiment 2 during week 0-2 and 3-4 respectively a FCR at 0.8 g dry matter intake (g gain)⁻¹

Period	Feeding rate %	Initial weight	Final Weight	Survival %	Weight Gain (%)	FCR g dry matter intake(g gain ⁻¹)	SGR
Week 0-2	8-8	1.09	3.27 ^b	100	300.9 ^d	0.69 ^d	7.88 ^d
	10-8	1.12	3.69 ^{ab}	100	330.8 ^c	0.72 ^d	8.54 ^c
	12-8	1.09	3.98 ^a	99	364.2 ^{ab}	0.77 ^c	9.23 ^{ab}
	14-8	1.08	4.06 ^a	100	377.9 ^a	0.85 ^b	9.50 ^a
	16-8	1.09	3.90 ^a	100	358.2 ^b	1.07 ^a	9.11 ^b
	Pooled SE	0.02	0.15		12.81	0.04	0.27
	P value	0.481	0.004		≤ 0.001	≤ 0.001	≤ 0.001
Week 3-4	6-6	3.27 ^b	6.65 ^b	100	203.5 ^b	0.65 ^d	5.07 ^b
	8-6	3.69 ^{ab}	8.55 ^a	100	231.7 ^a	0.70 ^d	6.00 ^a
	10-6	3.98 ^a	9.21 ^a	98	231.6 ^a	0.93 ^c	6.00 ^a
	12-6	4.06 ^a	9.26 ^a	100	227.7 ^a	1.23 ^b	5.88 ^a
	14-6	3.90 ^a	8.78 ^a	100	225.3 ^a	1.60 ^a	5.80 ^a
	Pooled SE	0.15	0.58		7.97	0.07	0.26
	P value	0.004	0.004		0.012	≤ 0.001	0.011
Week 0-4	8-8 and 6-6	1.09	6.65 ^b	100	612.4 ^b	0.66 ^d	6.48 ^b
	10-8 and 8-6	1.12	8.55 ^a	100	766.3 ^a	0.71 ^d	7.27 ^a
	12-8 and 10-6	1.09	9.21 ^a	99	843.7 ^a	0.88 ^c	7.61 ^a
	14-8 and 12-6	1.08	9.26 ^a	100	860.7 ^a	1.09 ^b	7.69 ^a
	16-8 and 14-6	1.09	8.78 ^a	100	807.0 ^a	1.41 ^a	7.46 ^a
	Pooled SE	0.02	0.58	0.00	49.29	0.06	0.24
	P value	0.481	0.004		0.001	≤ 0.001	0.001

FCR were significantly different among feeding treatment 3-5 and there were not considerably different between lesser feeding regime (feeding treatment 1 and 2) and these were significantly lower than excess feeding regime (feeding treatment 3-5) at the end of 1st two period (Table 11).

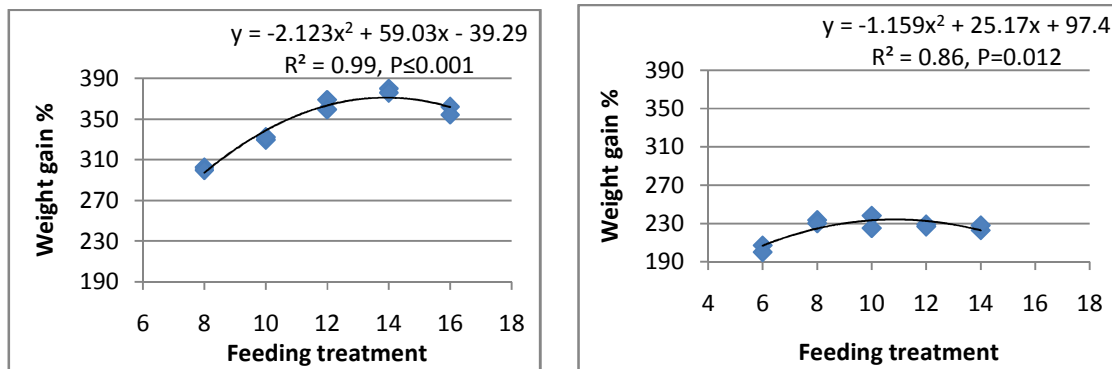


Figure 27. Weight gain% of experiment 3 during week 0-2 (left side) and 3-4 (right side) fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

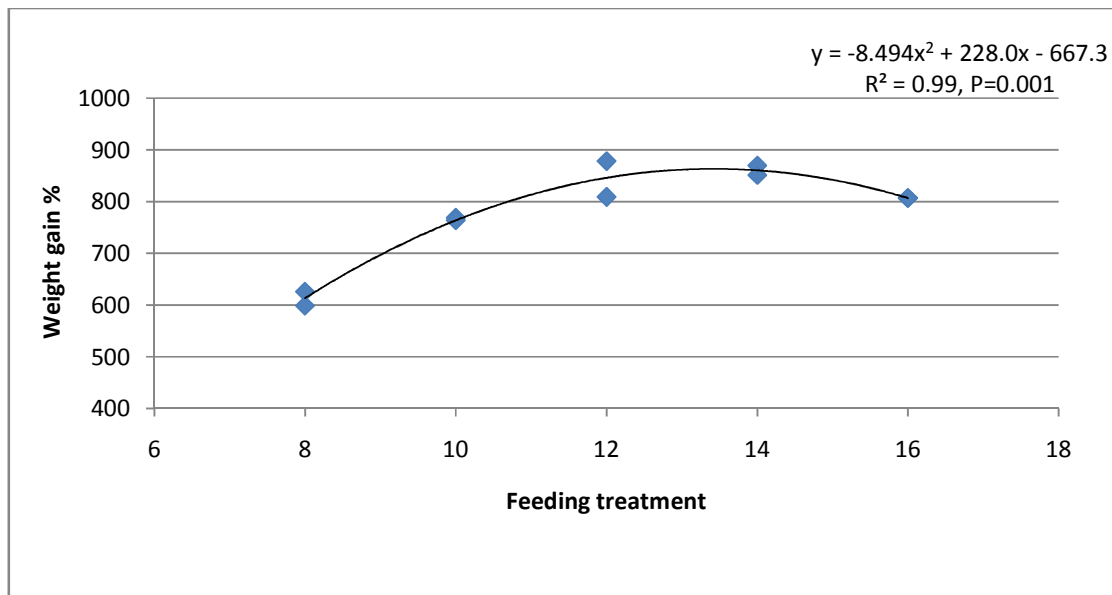


Figure 28. Weight gain% of experiment 3 during week 0-4 fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

At the end of last two week, fish weights were significantly differed ($P=0.004$, table 11). Weight gain% and SGR significantly higher at moderate to excess feeding regime (feeding treatment 2-5). FCR were considerable differed among feeding treatment 3-5 and not substantially different between feeding treatment 1 and 2. As a whole experiment, Table 11 and Figure 28 and 32 shows that highest weight gain% and SGR were achieved at feeding treatment moderate to excess feeding regime (feeding treatment 2-5). Meanwhile, FCR were significantly lower (0.66 to 0.71) at most lesser to moderate feeding treatment 1 to 2 while other feeding rate ranged from 0.88 to 1.41.

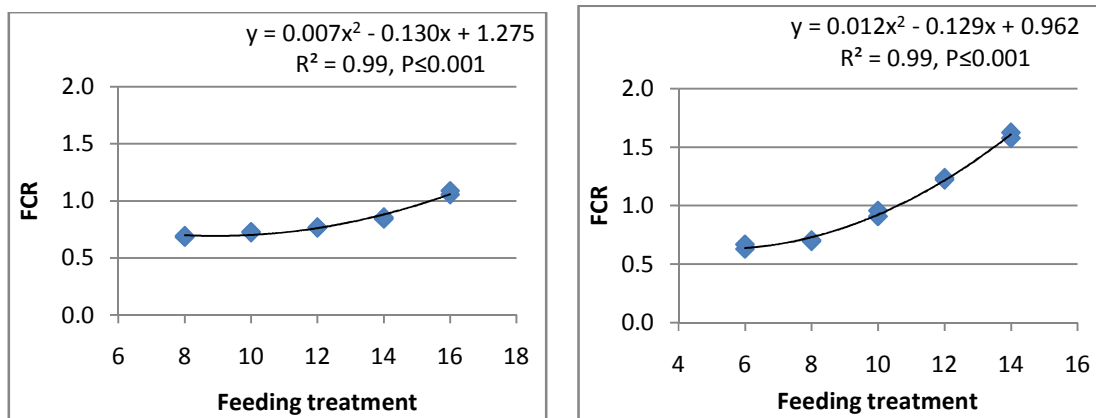


Figure 29. Feed conversion ratio (FCR) of experiment 3 during week 0-2 left side and week 3-4 right side fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

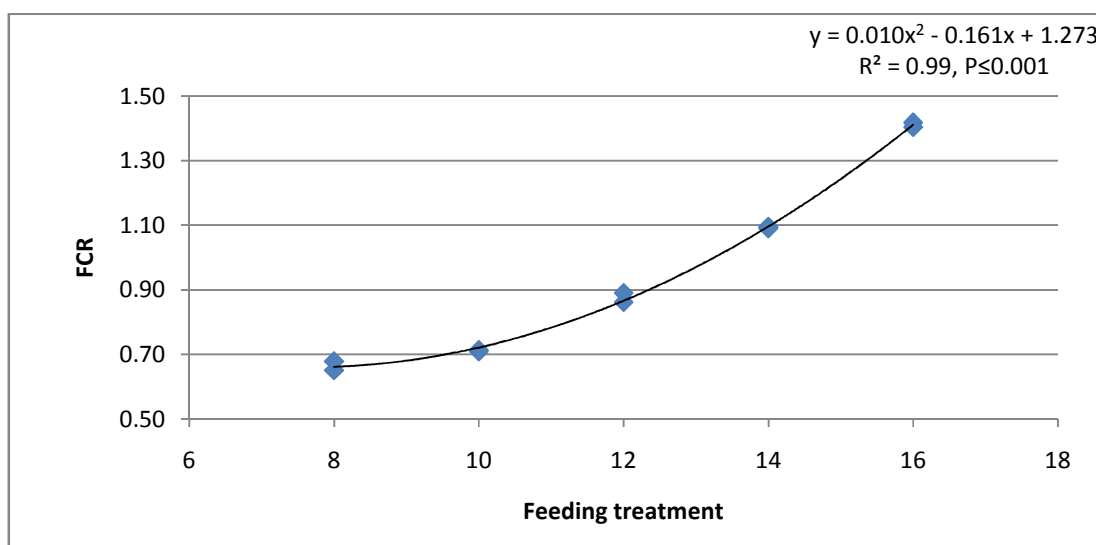


Figure 30. Feed conversion ratio (FCR) of experiment 3 during week 0-4 period fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

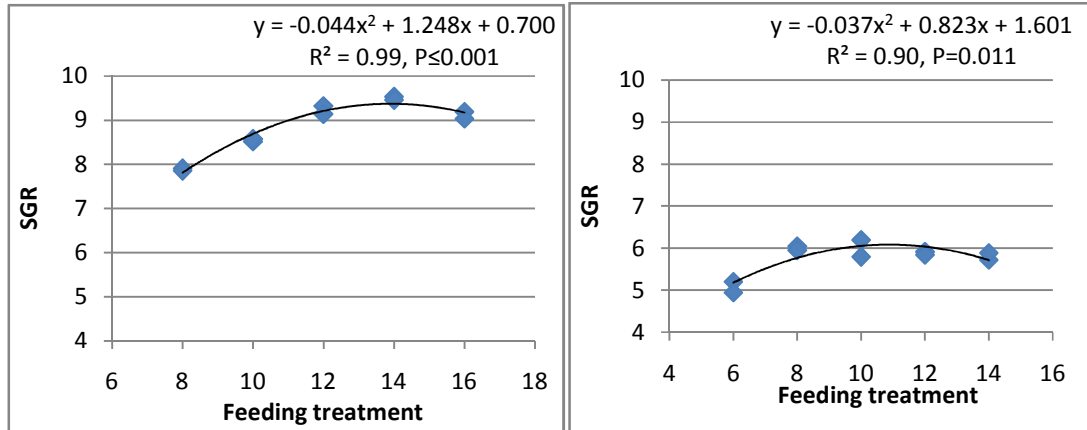


Figure 31. Specific growth rate (SGR) of experiment 3 during week 0-2 left side and week 3-4 right side fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

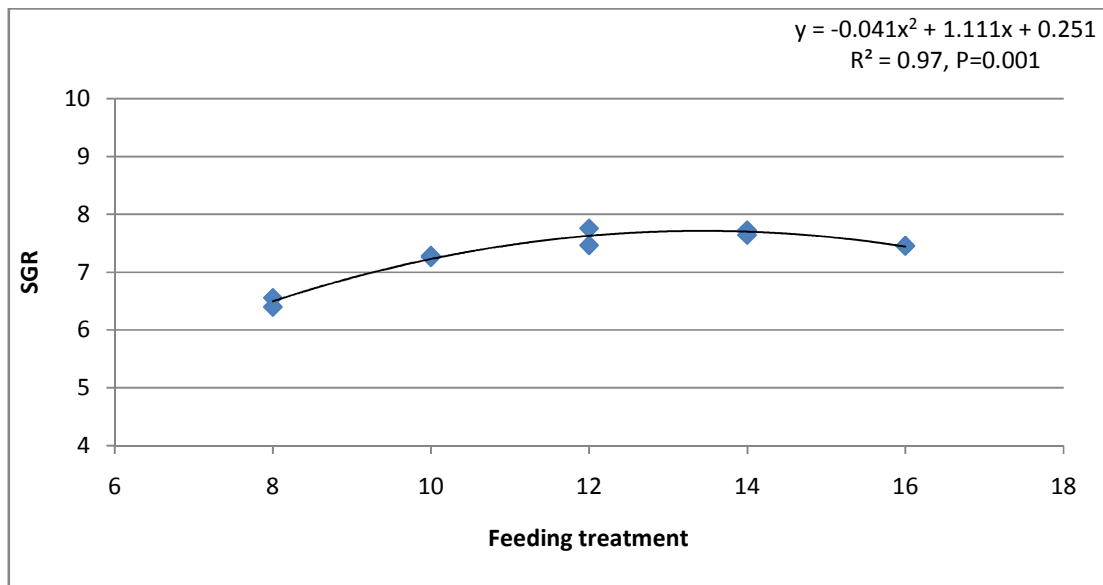


Figure 32. Specific growth rate (SGR) of experiment 3 during week 0-4 period fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

3.3.3 Body composition

Dry matter, crude protein and fat, ash and energy content of initial body composition were significantly differed by declining feeding rate treatment ($P \leq 0.001$, 0.002 , ≤ 0.001 , 0.005 and ≤ 0.001 respectively Table 12). Highest dry matter were found at treatment 3-4

while intermediate at top most feeding rate treatment 5 at the same time lesser feeding rate (Treatment 1 and 2) were significantly lower than excess feeding rate (Treatment 3-5). Crude protein contents were significantly higher at treatment 3-4, intermediate at treatment 2 and 5 and considerable lower at lesser feeding rate (treatment 1, Table 12). Crude fat were significantly higher (15.94 to 16.94%) at excess feeding rate (treatment 3-5), comparatively lower (11.50%) at moderate feeding rate (treatment 2) and intermediate at lowest feeding rate (treatment 1). Ash contents were likely to be significantly higher at lesser feeding regime (treatment 1 and 2), intermediate at excess feeding regime (treatment 3 and 5) and lower at treatment 4. Energy contents were significantly higher at excess feeding regime treatment 3-4 and intermediate at treatment 5 while significantly lesser at poorer feeding regime treatment 1 and 2 though clearly higher than initial energy contents (Table 12).

Table 12. Body composition of juvenile Nile tilapia Experiment 3, fed with declining rate 8 -16 to 8 % and 6-14 to 6%.

	Dry matter (%)	Crude Protein (%)	Crude Fat (%)	Ash (%)	Energy (MJkg ⁻¹)
Initial Body Composition	19.93 ^d	11.43 ^c	5.67 ^c	2.01 ^b	4.55 ^d
Final Body Composition					
Treatment 1 (8-8 and 6-6%)	26.71 ^{cd}	13.50 ^{bc}	8.13 ^{bc}	2.53 ^a	5.42 ^{cd}
Treatment 2 (10-8 and 8-6%)	29.22 ^{bc}	14.35 ^{abc}	11.50 ^b	2.41 ^a	7.41 ^{bc}
Treatment 3 (12-8 and 10-6%)	38.08 ^a	17.05 ^a	16.94 ^a	2.26 ^{ab}	10.12 ^a
Treatment 4 (14-8 and 12-6%)	38.0 ^a	17.36 ^a	16.7 ^a	2.02 ^b	9.9 ^a
Treatment 5 (16-8 and 14-6%)	35.70 ^{ab}	15.91 ^{ab}	15.94 ^a	2.22 ^{ab}	9.51 ^{ab}
Pooled SE	2.38	0.87	1.35	0.14	0.73
P value	≤0.001	0.002	≤0.001	0.005	≤0.001

3.3.4 Nutrients and energy retentions

Nitrogen, total phosphorus and energy retention were significantly affected by decline feeding rate treatment 1-5 ($P=0.001$, ≤ 0.000 and 0.004 Table 13) and Fig. 33 shows that Nitrogen and total phosphorus retentions were likely to be decreased as feeding rate proceed. Nitrogen retention significantly higher at low down feeding rate (treatment 1), intermediate at treatment 2 while excess feeding regime (treatment 3-5) were

significantly different with each other and poorer (41.2 to 74.8) compare to lesser feeding regime.

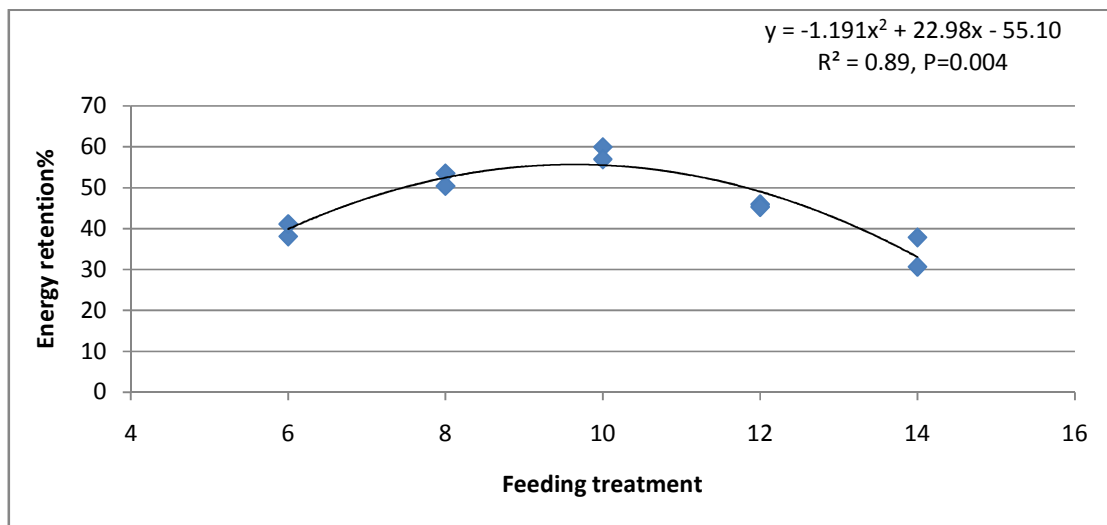
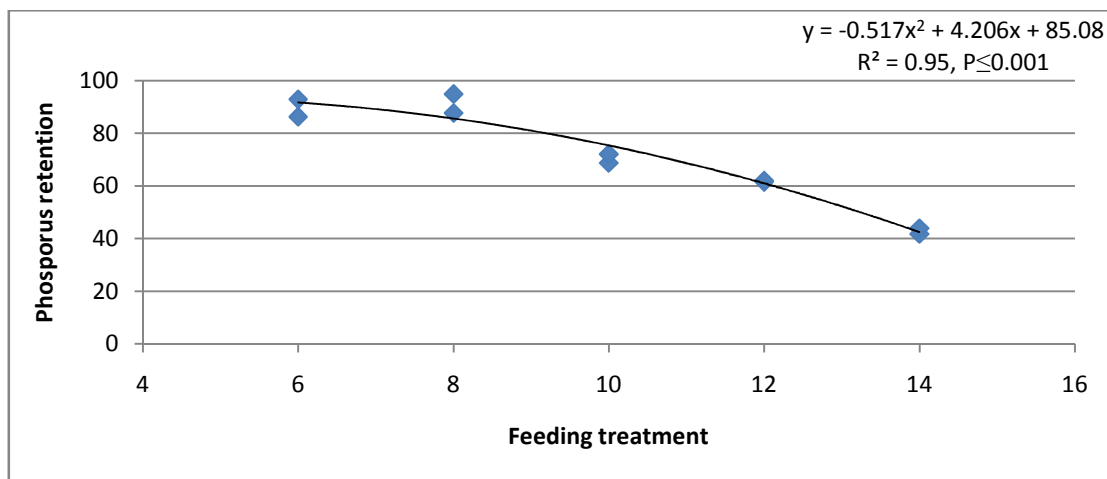
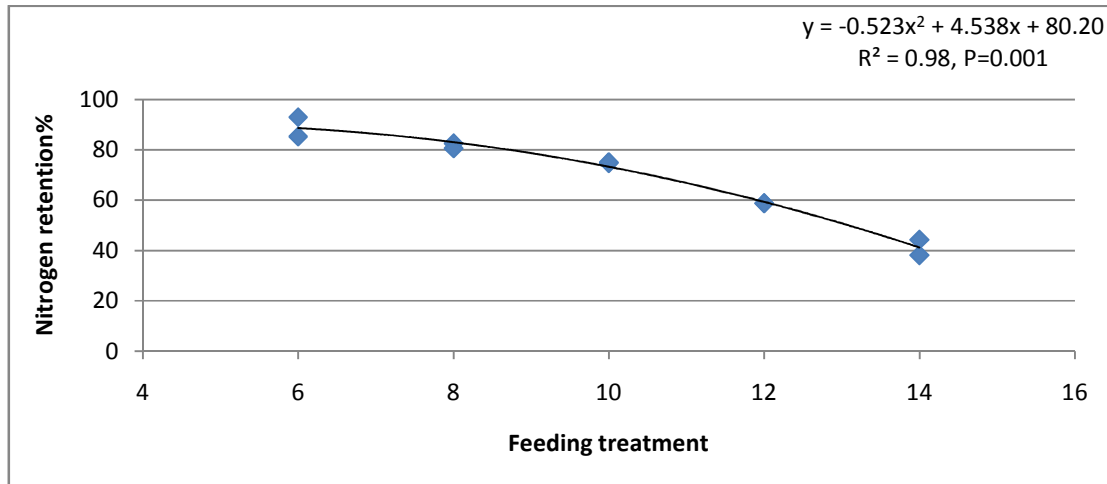


Figure 33. Nitrogen, phosphorus and energy retentions of experiment 3 fed with declining rate 8 -16 to 8 % (week 0-2) and 6-14 to 6% (week 3-4)

Total phosphorus retentions were significantly higher at lesser feeding regime (treatment 1-2) and not significantly differ with each other and lower retentions were found at excess feeding regime (treatment 3-4, Table 13). Energy retentions were considerably higher (58.5, Table 13) at treatment 3 than all other treatments of which were not significantly differed with each other.

Table 13. Nitrogen, phosphorus and energy retentions of juvenile Nile tilapia experiment 3 fed with declining rate 8 -16 to 8 % and 6-14 to 6% designed to check experiment 2.

Feeding treatment	Nitrogen retention (%)	Phosphorus retention (%)	Energy retention (%)
Treatment 1 (8-8 and 6-6%)	78.2 ^a	89.6 ^a	39.6 ^b
Treatment 2 (10-8 and 8-6%)	81.5 ^a	91.3 ^a	45.1 ^b
Treatment 3 (12-8 and 10-6%)	74.8 ^b	70.5 ^b	58.5 ^a
Treatment 4 (14-8 and 12-6%)	58.7 ^c	61.8 ^b	45.6 ^b
Treatment 5 (16-8 and 14-6%)	41.2 ^d	42.9 ^c	34.3 ^b
Pooled SE	3.56	4.54	6.81
P value	0.001	≤0.001	0.004

3.4. Experiment 4

Fishes (77.9±0.03 g) were fed at five different feeding rate 1, 2, 3, 4 and 5% body weight per day) expected a FCR at 9.0 g dry matter intake gain⁻¹.

3.4.1 Water quality

The temperature and pH were found as 25.6±0.5°C and 6.8±0.1 respectively during whole experiment (total 13 observations). Oxygen, TAN and NO₂ concentrations were significantly influence by feeding rate 1-5% (P=0.005, 0.011 and 0.005 respectively, Fig 34). Oxygen concentration were decreased as feeding rate increased while opposite trend were found for TAN and NO₂. Lowest and highest TAN content were found at 1 % (0.28±0.03 mg l⁻¹) and 5% (1.0±0.00 mg l⁻¹) feeding level respectively. On the other hand lowest and highest NO₂ were found at 1 % (0.6±0.00 mg l⁻¹) and 5% (0.1±0.00 mg l⁻¹), respectively.

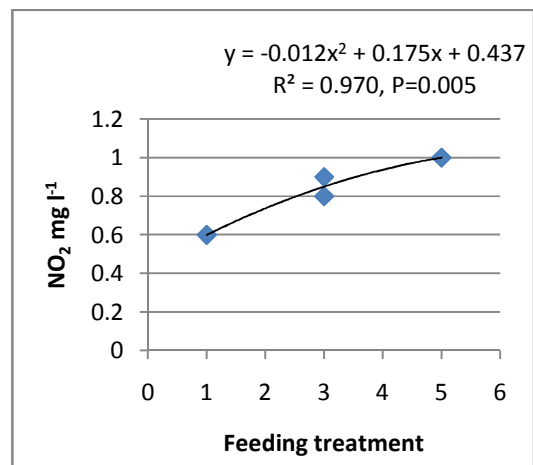
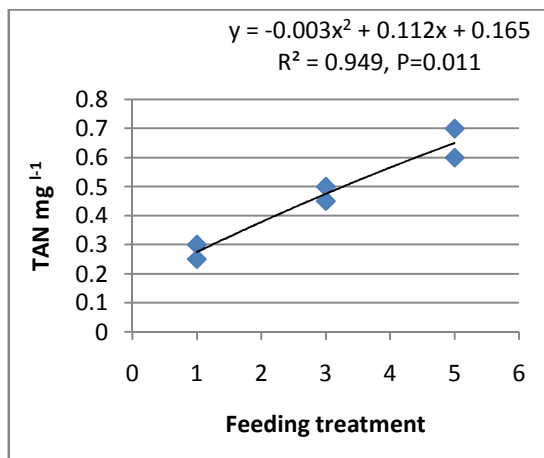
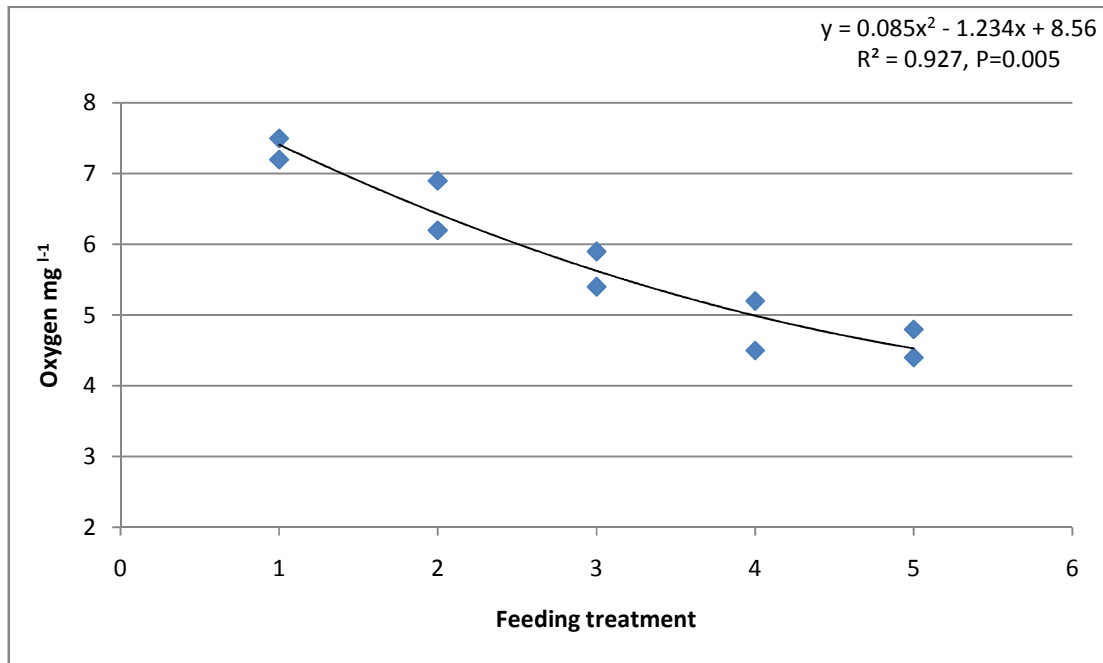


Figure 34. Oxygen, TAN and NO₂ concentration of Expt. 4 fed with 1-5% body weight gain expected FCR 0.8 g dry matter intake (g gain⁻¹)

3.4.2 Growth parameter

Initial weight of Nile tilapia of experiment 4 were not significantly differed ($P=0.805$). At the end of two week fish weight, weight gain%, FCR and SGR were significantly differed

by given feeding regime ($P \leq 0.001$) and all fish were survived (Table-14, Figure 35 and 36). Weight gain% and SGR were significantly higher at excess feeding regime (3-5%) than lesser feeding regime (1-2%) and there were significantly different between 1 and 2% feeding rate. FCR were ranged from 0.86 to 1.75 (Table 14) in which lesser to moderate feeding regime (1-3%) was significantly lower than excess feeding regime (4-5%). And there were significantly higher at excess feeding rate (4-5%) and differed between these.

Table 14. Growth performance of adult Nile tilapia Expt. 4 fed with 1-5% body weight expected FCR 0.8 g dry matter intake (g gain^{-1})

Period	Feeding rate %	Initial weight	Final Weight	Survival %	Weight Gain (%)	FCR g dry matter intake(g gain^{-1})	SGR
Week 0-2	1	77.9	88.6 ^c	100	113.8 ^c	0.89 ^c	0.99 ^c
	2	77.9	101.5 ^b	100	130.3 ^b	0.86 ^c	2.03 ^b
	3	77.9	114.9 ^a	100	147.5 ^a	0.87 ^c	2.99 ^a
	4	77.9	115.1 ^a	100	147.7 ^a	1.26 ^b	3.00 ^a
	5	77.9	113.9 ^a	100	146.1 ^a	1.75 ^a	2.92 ^a
	Pooled SE	0.03	4.31		± 8.5	0.14	0.33
	P value	0.805	≤ 0.001		≤ 0.001	≤ 0.001	≤ 0.001

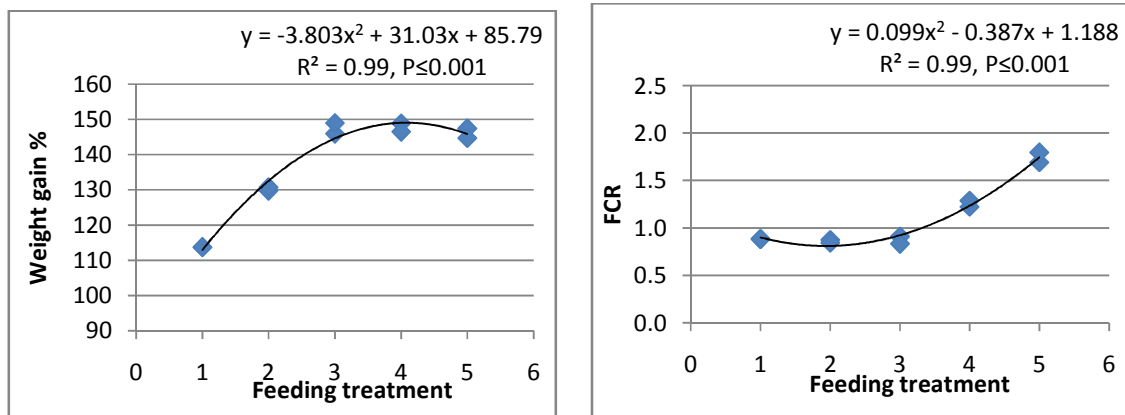


Figure 35. Weight gain % (left side) and feed conversion ratio (right side) of Expt. 4 fed with 1-5% body weight gain expected FCR 0.8 g dry matter intake (g gain^{-1}).

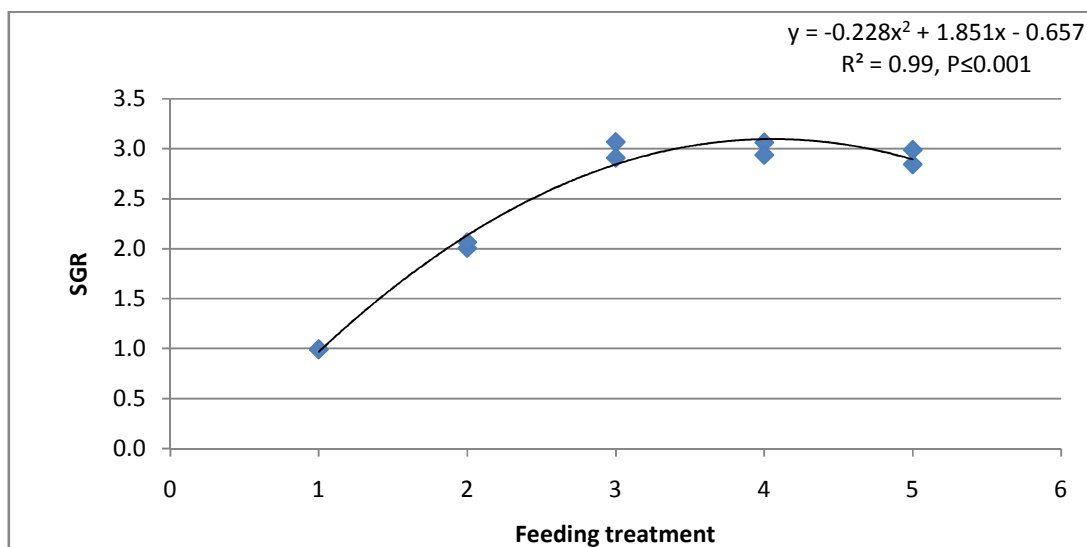


Figure 36. Specific growth rate of experiment 4 fed with 1-5% body weight gain expected FCR 0.8 g dry matter intake (g gain^{-1}).

3.4.3 Body composition

Feeding rate 1-5% body weight of Expt.4 was significantly affected to dry matter, ash and energy contents of tilapia. Significantly higher dry matter content were found at 4% feeding rate while intermediate at 3% and 5% feeding rate. Ash contents were significantly decreases with increasing feeding rate (Table 15). Energy contents were considerably higher at feeding rate 3-5% and intermediate at 2% feeding rate. Crude proteins were significantly affected by given feeding resume.

Table 15. Body composition of adult Nile tilapia fed with 1-5% body weight of experiment 4 expected FCR 0.8 g dry matter intake (g gain^{-1})

Fish	Dry matter (%)	Crude Protein (%)	Crude Fat (%)	Ash (%)	Energy (KJg^{-1})
Initial Body Composition	27.88 ^c	15.7	7.43	3.91 ^a	6.61 ^b
Final Body Composition					
1%	28.25 ^{bc}	15.85	8.74	3.78 ^{ab}	6.55 ^b
2%	28.72 ^{bc}	15.58	8.82	3.45 ^{abc}	7.09 ^{ab}
3%	30.33 ^{ab}	15.11	10.84	3.29 ^{bc}	7.75 ^a
4%	31.27 ^a	15.52	11.00	3.34 ^{bc}	8.00 ^a
5%	30.24 ^{abc}	15.37	10.4	3.15 ^c	7.78 ^a
Pooled SE	0.51	0.06	0.65	0.09	0.20
P value	0.008	0.077	≤ 0.05	0.010	0.003

3.4.4 Nutrient and energy retentions.

Nitrogen retentions were significantly higher at limited to moderate feeding rate (1-3%, $P \leq 0.001$) and considerable lower at excess feeding rate (4-5%, Table 16). Energy retentions were significantly higher at 3%, intermediate at 2% feeding rate and significantly lower at 1, 4 and 5% feeding rate ($P=0.001$).

Table 16. Nitrogen and energy retention of adult Nile tilapia fed with 1-5% body weight of experiment 4 (start weight 77.9 ± 0.3).

Feeding treatment	Nitrogen retention (%)	Energy retention (%)
1%	49.41 ^a	32.3 ^{cd}
2%	45.28 ^a	46.9 ^{ab}
3%	40.89 ^a	54.31 ^a
4%	30.98 ^b	40.58 ^{bc}
5%	21.59 ^b	27.64 ^d
Pooled SE	2.18	7.92
P value	≤ 0.001	0.001

NB: Phosphorus retention were found to be error on measurement and subjected to analyze again.

CHAPTER FOUR

4: Discussion

The present study clearly demonstrated that feeding rates significantly affected the water quality parameters, growth performance, body composition, nutrient and energy retentions except the adult tilapia (≥ 257.9 g). The apparent crude protein digestibility was significantly affected by feeding rate in adult tilapia. In Expt.4, report show that crude protein and fat content of body composition were not significantly affected by feeding rate (1-5%) in tilapia (≥ 77.9 g)

4.1 Effect of feeding rate on water quality parameters

The temperature of four experiment (average 25.5 to 26.7⁰C) were almost same as like experiment on effect of stocking density on water quality and production of red tilapia in recirculated water system (Suresh and Lin, 1992). It has been noted that temperature 26-28⁰C optimum for 10-1000 g Nile tilapia growth (Bergheim, 2007). pH (6.8) was also within acceptable limits (6.5 to 7) according to finding of water quality criteria for recirculation systems (Bergheim, 2007) during whole experiment period.

Dissolved oxygen content of all four experiment were found to decrease as the feeding level increase while TAN and NO₂ increase as the feeding level in range from. As a result feeding treatment significantly influence on water quality in the rearing tank. It may be increasing feeding rate would add to feces in the water thus influence to decrease oxygen and increase TAN and NO₂ concentration in the rearing tank. (Singh et al., 1999) found that increasing feeding rate significantly influences TAN but NO₂ did not show any significant effect. Despite increasing TAN and NO₂ and decreasing oxygen concentration in the rearing tank water due to increase the feeding rate, still mentioned parameters remained acceptable limit. Recommendation level for oxygen is above 3 mg l⁻¹, TAN 0.5 to 1.0 mg l⁻¹, (Chapman, 1992) and NO₂ below 1.0 mg l⁻¹ (Rakocy, 1989). It may be the end biomass of four experiments were not exceed as a overstocking. The present showed those maximum yields were obtained 4.7 kg/210 liter and 2.3 kg/115

liter in big and small tank, respectively. It is being supported that lower density fish (3.75 kg m^{-3}) grew significantly better (Suresh and Lin, 1992).

4.2 Effect feeding rate on survival.

Feeding rate did not influence any mortality in any of the experiments. Thus, the results do not support previous findings that juvenile tilapia is more sensitive to feeding rate than larger tilapia. The reason may be that restrictions in feeding rate were not enough for feed triggering cannibalism behavior. Macintosh and De Silva (1984) found that cannibalism was inversely related with feeding rate in hybrid tilapia fry. Santiago et al. (1987) found that tilapia smaller than 12 mg were fed at 15 to 60% feeding rate and found that survival rates were increased up to (87%) at feeding rate 45% then decreased. Feeding rate (10-35%) in average weight of 0.016 g tilapia showed increased survival rate with increasing feeding rate (El-Sayed, 2002). The combination of fish density and high feeding rates was not sufficient to deteriorate water quality to a level where the tilapia suffered notable health problems.

4.3. Effect of feeding rate on growth parameter, body composition, nutrient retentions and protein digestibility of Expt. 1

The feeding rate and frequency in relation with feed consumed and the efficiency are prime factors in determining growth rate. There is a positive relation between growth and feeding frequency and satiation feeding of tilapia at 4-h intervals should increase production efficiency (Riche et al., 2004b). Moreover, author also explained tilapia feed three times per day give better result in terms of growth and efficiency. In experiment 1, effect of feeding rates does not give significance effect on weight gain% and FCR but SGR were affected only during week 0-3 period though WG% and FCR increased linearly where highest SGR were obtained at 115% (1.8 ± 0.0) feeding level were likely to be higher than all other feeding treatment.

This growth results obtained at the end of week 0-3 (379.5 ± 1.3 g) were higher compared to result obtained by (Garduño-Lugo et al., 2003). They compared growth in Nile and red tilapia and found that fish fed a commercial diet (Moisture $10.9 \pm 6.96\%$; crude protein, $36.8 \pm 3.37\%$; crude lipid, $14.1 \pm 5.23\%$; ash, $7.52 \pm 0.68\%$) having initial (139.3 g) and final weigh (384.4 g) and SGR 1.04 for Nile tilapia.

Growth rates and FCR were not significantly different from the results obtained by Garduño-Lugo et al. (2003). Present study showed similar result during week 4-6 and considering the whole experiment period. Since there were uneaten feed and available egg observed during daily flushing the tank water which means feeding rate has great influence on sexual maturity in last two weeks.

When the Nile tilapias get ready to spawn they stop eating and due to mouth breeder behavior, they keep egg until hatch out. Other reason is that longer photoperiod (18L: 6D) can improve spawning synchrony (Campos-Mendoza et al., 2004). They also mentioned that there were not significance differences in weight gain (mean final weight ranged from 477 ± 40 to 577 ± 40) under different lighting period. Gunasekera et al. (1996) found that 35% crude protein diet can significantly improve larval quality and performance in Nile tilapia. Therefore, it shows that this experiment synchronized spawning due to longer photoperiod (24h L) and crude protein content of diet. As whole experiment, it reveals that feeding rate did not significantly influence on the weight gain%, FCR and SGR. As a result, feeding rate is likely to be proficient at restricted feeding rate 55% (1.24 g /day).

Similar study with different feeding rate support this study where maximum growth was achieved at feeding rate near satiation while feed conversation ratio improved at lower feeding rate (Clark et al., 1990) . This study result shows that FCR (1.01) feeding rate at 1.24% body weight/day is better than feeding rate at 2.42% body weight/day having FCR 1.57 studies with feeding rate ranged 2.42 to 3.56%/day (Clark et al., 1990). Another study on improvement of rapeseed meal through addition of organic salt found

that FCR 1.49 where start and final weight (224 and 351.3 respectively) of fish (Gao, 2011).

Body composition of Nile tilapia Expt.1 was not changed by feeding regime except the energy contents ($r^2=0.81$, $P=0.036$) and fat contents likely to be tending at restricted feeding rate (55%, $P=0.066$). The energy contents were significantly higher at excess feeding regime (100-115%) and intermediate at restricted feeding regime (55-85%) compared to initial energy contents. The reason is that the tilapias are able to store lipid in the carcass and viscera but not able to utilized this source for growth (Hanley, 1991).

Nitrogen retention, phosphorus and energy retention were decreased approximately linearly as the feeding rate increased though feeding regime does not sway on phosphorus and energy retention but nitrogen retention were significantly affected by feeding regime. Result (Table 7) show that significantly higher at 55% (42.6%) feeding rate which is similar study with 25% fish protein diet feeding rate at 2.5% (Viola et al., 1988). Energy retention of Expt.1 were higher 52.2% which is likely to intended at 55% feeding rate ($P=0.065$) that also higher than study carried out with extruded rapeseed meal (Gao, 2011).

An apparent crude protein digestibility of extruded diet of this experiment increased as the feeding rate proceeded till 100%. The values obtained were found slightly higher than obtained with a diet with extruded rapeseed meal (Gao, 2011). Digestibility of this study was hypothesized to be higher at restricted feeding rate because proportion of enzyme secretion might be higher in the gut compared to amount of ingested feed. However, the present finding may indicate that the relative contribution of endogenous nitrogen from the gut to the feces may have been stable, while proportion of not digested nitrogen from the feed increased. The present study of ACPD trend is line of juvenile grass carp where lower digestibility were found at restricted feeding rate than increased up to certain level afterward decreased (Du et al., 2006).

4.4 Effect of feeding rate on growth performance, body composition, and nutrient retention of juvenile Nile tilapia of Expt. 2.

Growth performance of juvenile Nile tilapia shows that weight gain% were not same between 1st and 2nd two week though it was significantly affected by feeding regime in Expt. 2. It might be reason that weight gain relatively decrease as the body weight of fish increase (Xie et al., 1997a). Weight gains were increased up to 100% feeding rate than start to fall down. It discloses that over feeding does not support for the growth. It support the other study where fish fed with higher than optimum feeding do not necessarily benefit from excess feed (Abdelghany and Ahmad, 2002). It may be reason that apparent digestibility decreased with increasing feeding rate (El-Saidy et al., 2005) and but present finding from Expt. 1. (Fig. 16) indicate that apparent crude protein digestibility decreased in excess feeding rate (115%). Therefore, it can be recommended that production can be maximized through using limited feed amount.

Table 8 indicate that maximum growth were achieved at 85% feeding rate during whole experiment period that mean 6.8% and 5.1% body weight feeding per day were suitable for maximum growth of juvenile (1.1 g) during 1st and 2nd two week. At the same time, SGR at 7.19 were achieved at same feeding treatment. FCR were relatively higher in 2nd two week comparatively than 1st two week in all feeding treatments (Table 8, Fig 21). It may be requirement of body maintenance energy is relatively higher as the juvenile grow out and energy may be used for body maintenance rather than growth. Same suggestion were found (Ali et al., 2008) study with feeding different protein to energy ratio and effect of growth and body composition of Nile tilapia fingerlings. Result (Table 8) of this study shows that lower FCR were found at restricted feeding regime (55-85%) and significantly higher at excess feeding regime (100-115%) in both 1st and 2nd two week. It seems to be proficient at 85% feeding rate (FCR 0.81) where highest weight gain% and SGR were obtained. Recent study using 13.5% fish meal working with less than 2 g of juvenile tilapia for 8 week (Zhao et al., 2010) shows that FCR (0.95) and SGR (6.27) are give the impression that using 7% fish meal of present study would to be expected proficient.

Body composition of dry matter, crude protein, fat, ash and energy contents of whole body of juvenile Nile tilapia were significantly affected by experimental feeding regime. Dry matter and crude fat contents were lower at 55% and higher at 115% feeding rate (Table 9). It means that with increasing feeding rate, dry matter and crude fat add to the body composition though there were some variations at 85-100% feeding rate and reason is unknown. However present trend is not line with El-Saidy et al. (2005) where feeding rate does not significantly influence on crude fat but it is on procession in which dry matter and fat content are increased with feeding rate studied with juvenile Chinese sucker (Yuan et al., 2010). They explained that fat content were not significant difference in excess feeding regime which is similar result with present finding. Crude protein content of juvenile tilapia showed negative trend as the feeding rates increased, in contrast to the findings of Yuan et al. (2010) that protein contents were increased with feeding rate. There were similar trend found study with feeding rate on adult tilapia (El-Saidy et al., 2005). Ash contents were like to be not significance different among feeding treatment and it's also like same as El-Saidy et al. (2005) finding. Energy contents of whole body juvenile Nile tilapia shows (Table 9) that it increased up to certain level than decreased at excess feeding rate (115%) and significantly lower at restricted (55%) feeding rate but others were likely to be not significant difference but El-Saidy et al. (2005) found no significant different with different feeding rate.

Nitrogen, phosphorus and energy retentions were significantly higher at restricted feeding regime. It means that nutrient and energy deposition by digestion is more efficient at restricted feeding rate for juvenile Tilapia. Same observation have been reported from the rainbow trout (Bureau et al., 2006) and protein retention of juvenile grass carp were also higher at lower feeding regime (Du et al., 2006). But opposite observation were found in juvenile of Indian catfish (*Heteropneustes fossilis*) where protein and energy retention were lower at restricted feeding regime (Ahmed, 2010).

4.5 Effect of feeding rate on growth performance, body composition, and nutrient retention of juvenile Nile tilapia of Expt. 3.

Declining feeding rate of Expt.3 were designed to compare the growth performance, body composition and nutrients retention of feeding rate using 8% and 6% body weight at 100% level in Expt.2. Initial weight were not significant different between Expt.2 and Expt.3. Weight gain%, SGR and FCR were significantly influenced by different declining feeding regime in both 1st and 2nd two week. Weight gain%, SGR and FCR of this study treatment 1 (8 & 6%) were lower (Table 11) than 100% feeding rate of Expt. 2 (Table 8). It may be reason that water qualities of both experiments were not same. According to Fig. 18 and Fig. 26, TAN and NO₂ contents of Expt. 3 were little bit higher than Expt. 2 at all experimental feeding treatment and these parameters were acceptable limit. Though there were almost same initial biomass but TAN and NO₂ concentration of Expt.3 revealed that there were might be error in recirculation system where nitrification process might be hindered.

Considering whole Expt. 3, Weight gain% and SGR were significantly higher at treatment 2 to 5 and significant different among these treatments. It suggest that treatment 10-8 and 8-6% feeding rate is adequate in order to get higher weigh gain and SGR meanwhile FCR (0.71) would be significantly lower than excess feeding regime. It implies that declining feeding rate is better in terms of weight gain, SGR and FCR compare to fixe feeding rate (Expt.2) for juvenile Nile tilapia. Body composition of dry matter, crude protein, fat and energy contents were significantly affected by declining feeding rate (Table. 12) and it shows that dry matter, crude protein and fat increased (except ash) with increasing feeding rate and decreased at excess feeding rate. It recommend that body composition of treatment 3 (12-8 & 10-6%) were adept to be better taking into consideration than Expt. 2.

Nitrogen and phosphorus retention were significantly higher at lower feeding rate but compare to Expt. 2 where 100% feeding rate were lower nutrient and energy retention. Treatment 1 of Expt. 3 and 100% feeding rate of Expt. 2 in which same feeding rates was applied and reason is not clear. However, 8-8 & 6-6% feeding rate would be better in

terms of nitrogen and phosphorus retention but considering energy retention 12-8 & 10-6% feeding rate would be better compare to Expt.2

4.6 Effect of feeding rate on growth performance, body composition, nitrogen and energy retention of Expt.4

Growth performance includes weight gain%, SGR and FCR were significantly affected by feeding treatment (1 to 5% body weight, Table 14). It indicates that higher weight gain and SGR would achieved by implying 3% body weight feeding rate where efficient FCR (0.87) were also found. It also indicates that 1% feeding rate (FCR were higher than 2% feeding rate) was just above body maintenance level and still remain positive growth for 77.9±0.3 g adult tilapia. El-Saidy et al. (2005) suggest that 2% feeding rate is cost effective for Nile tilapia (60.7 to 221.1) in concrete tank. According to the finding, they showed that FCR and SGR were 2.5 and 0.66 respectively for 28 week period. Finding from GST 18 generation (Gjøen and Zimmermann, unpublished) also suggest for 2% feeding rate with expected FCR 1 in earthen pond condition. Present studies represent lower FCR (0.87) and higher SGR (2.99) for two week period. It suggests from all four experiments that growth rate decreases as the body weight increased. Same report were found study with different age group of tilapia due to decreased relative feed intake (Xie et al., 1997a). However, present study indicates that 3% feeding rate is like to be better at this stage for tilapia using every one hour meal. Moreover further research is needed using same condition in pond environment.

Crude protein and fat does not significantly affected by feeding treatment but dry matter, ash and energy content significantly affected by provided feeding regime. It means that tilapia larger than 77.9 g feeding rate does not affect crude protein and fat. Same reports were found from Expt. 1. Dry matter was significantly higher at 4% while intermediate at 3 and 5% feeding rate. It revealed that increasing feeding rate can increase dry matter content up to certain level but further increasing feeding rate does not support for dry matter improvement. The statement also supported from previous experiment with juvenile tilapia. But study from El-Saidy et al. (2005) shows that moisture content does

not significantly affected by different feeding rate. It might be reason that smaller tilapia are able increase dry matter content in their body compare to larger tilapia. This proclamation is also supported by Expt.1 where dry matter content did not significantly differed by feeding rate for larger tilapia. Ash content significantly reduced compare to initial body composition. Result from Expt.1 also reduced as feeding rate increases though ash contents were not significantly affected by feeding rate. It point out that lower the feeding rate better energy contents in tilapia fish body for larger tilapia. But study from El-Saidy et al. (2005) doesn't agree with this observation. Energy contents significantly were higher at 3-5% feeding rate while intermediate at 2% feeding rate. Same trend line was also found from Expt.1 which put forward that increasing feeding rate can enhance the energy content of larger tilapia. In case of juvenile tilapia energy content can be increased with increasing feeding rate at certain stage but further increased feeding rate doesn't support for energy add to body composition. It might be reason that tilapia are to utilize better energy at lower feeding rate. Same trend were found study using 0.5 to 4% ration level having average initial weight 8.29 to 11.2 (Xie et al., 1997b).

Nitrogen retentions were significantly higher at 1-3% feeding rate than excess feeding regime (4-5%). Nile tilapia feeding with fish meal and replacing plant protein showed that found that nitrogen balance 431 to 480 g/kg-1 which is almost similar to present study (40.89 to 49.41%) at lesser to 3% feeding rate (Schneider et al., 2004). It gives suggestion from this study that lower the feeding ration better nitrogen utilization and juvenile tilapia is more efficiently utilize nitrogen than larger tilapia. Energy retention increasing with increasing feeding ration up to 3% feeding rate but further increasing doesn't sustain for energy retention.

Conclusion

In this study, it concludes that growth and feed utilization of genetically improved Nile tilapia can be maximized using correct feeding rate. Optimal feeding rate of juvenile tilapia is higher than that of larger tilapia as the fish growth relative feed intake decrease. It is also abridged that growth can be hampered due to sexual maturation. The present study suggest that declining feeding rate can be suitable for juvenile tilapia (1.1 g), and that daily feeding rates at 10-8% and 8-6% can be appropriate for 1st and 2nd two week periods, respectively. A feeding rate at 3% seems suitable for tilapia between 80 and 115 g, while fish larger than 260 g had best response to a feeding rate of 1.2% of body weight.

CHAPTER FIVE

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Appendix1. Layout of recycled fresh water tanks used in research

