



# International Journal of Fisheries and Aquatic Studies

E-ISSN: 2347-5129  
P-ISSN: 2394-0506  
(ICV-Poland) Impact Value: 5.62  
(GIF) Impact Factor: 0.549  
IJFAS 2017; 5(1): 149-156  
© 2017 IJFAS  
www.fisheriesjournal.com  
Received: 23-11-2016  
Accepted: 24-12-2016

**Rakesh Prasad Bhagat**  
Department of Zoology, Tri-  
Chandra Multiple Campus,  
Tribhuvan University,  
Kathmandu, Nepal  
e-mail: rakesh.bhagat2009@gmail.com

**Sudip Barat**  
Aquaculture and Limnology  
Research Unit, Department of  
Zoology, University of North  
Bengal, Siliguri, District,  
Darjeeling, West Bengal, India  
e-mail: sudipbarat@rediffmail.com

## Effect of artificial feed on survival and growth of rainbow trout, *Oncorhynchus mykiss* (Walbaum) during exogenous feeding in the raceways of Kathmandu, Nepal

Rakesh Prasad Bhagat and Sudip Barat

### Abstract

Low cost, alternative, and unconventional animal protein in replacement to costly shrimp meals were formulated in the artificial feed and their effect on survival and growth of rainbow trout, *Oncorhynchus mykiss* (Walbaum) during exogenous feeding in farmer's raceways were compared. Three feed formulations, two with animal protein of silkworm pupae (Treatment-1) and silkworm moths (Treatment-2) and one with synthetic amino acids of lysine and methionine (Treatment-3) were evaluated through survival and growth along with feed efficiency indicators against shrimp meals acting as control (Treatment-4). All the four diets (three formulated and one control) were fed to the free swimming fries, fries, and fingerlings for 150 days (5 months). There was a significant difference on survival ( $P < 0.01$ ) and growth ( $P < 0.01$ ) of above mentioned stages with all the diets. Survival rate with shrimp meals diet fed stages revealed superiority ( $P < 0.01$ ) over other feed formulations, silkworm pupae diet lesser survival, silkworm moths diet less survival, and synthetic amino acids diet least survival. However, survival due to shrimp meals diet was insignificant ( $P > 0.05$ ) with that of silkworm pupae diet. Growth with silkworm pupae diet fed stages showed superiority ( $P < 0.01$ ) over all the diets and synthetic amino acids diet lowest whereas shrimp meals diet and silkworm moths diet exhibited low and lower growth respectively. However, growth due to silkworm pupae diet was highly significant ( $P < 0.01$ ) with that of shrimp meals diet. So, total feed intake, total protein intake, and feed efficiency indicators of feed efficiency, protein efficiency ratio, absolute growth rate, specific growth rate, and relative growth rate were highest due to silkworm pupae diet, higher due to shrimp meals diet, high due to silkworm moths diet, and lowest due to synthetic amino acids diet. However, condition factor was highest due to silkworm pupae diet, higher due to shrimp meals diet, low due to synthetic amino acids diet and lowest due to silkworm moths diet. Unlike other feed indicators, feed conversion ratio exhibited highest due to synthetic amino acids diet, higher due to silkworm moths diet, low due to shrimp meals diet, and lowest due to silkworm pupae diet. Further, protein productive value percentage resembled crude proteins percentage present in the diets. The highest growth period was observed during May and lowest during December to January in all the diets during 2011 and 2012. Cost analyses revealed silkworm moths diet cheapest with low production cost, silkworm pupae diet cheaper with lowest, synthetic amino acids diet cheap with highest, and shrimp meals diet costly with high production cost. Results also indicated that natural and animal protein of silkworm pupae diet, shrimp meals diet, and silkworm moths diet was superior to synthetic amino acids diet. The results finally confirmed that cost effective silkworm pupae diet could be used as better alternative to completely replaced shrimp meals diet without compromising survival and growth.

**Keywords:** Formulated diets, rainbow trout, exogenous feeding, survival and growth

### 1. Introduction

Free swimming fries (FSFs) of rainbow trout, *Oncorhynchus mykiss* (Walbaum, 1792) start exogenous feeding when their yolk-sacs are completely absorbed (Pradhan *et al.*, 2008) [24]. During exogenous feeding, FSFs feed on natural feed. However, hatchery-grown FSFs are exclusively dependent on artificial feed for their survival and growth (Bardach *et al.*, 1972) [6]. FSFs of rainbow trout are totally dependent up on quality and quantity of artificial feed (Rai *et al.*, 2008) [25] that change 0.07-0.10 g FSFs into 200-300 g table fish in 16-18 months in the raceways of Kathmandu, Nepal (Anonymous, 2003) [3]. Therefore, corresponding increases in nutrition, feedstuffs, and feeding of artificial feed increase rainbow trout production.

### Correspondence

**Sudip Barat**  
Aquaculture and Limnology  
Research Unit, Department of  
Zoology, University of North  
Bengal, Siliguri, District,  
Darjeeling, West Bengal, India  
e-mail: sudipbarat@rediffmail.com

Artificial feed of rainbow trout should contain proteins (animal and plant) 40-50% (Robinson and Li, 1996) [27], carbohydrates 15-25% (Hasan, 2001) [15], lipids 10-15% (Robinson and Li, 1996) [27], minerals 1% (Hasan, 2001) [15], and vitamins 1% (Hasan, 2001) [15]. Artificial feed has been considered as the single largest operating cost in rainbow trout farming throughout the world including Nepal. Artificial feed alone is 76% of the total variable cost and 40% of the total production cost of rainbow trout culture (Nepal *et al.*, 2002) [22] and is one of the major constraints after seed supply to limit expansion of rainbow trout cultivation in Nepal. Therefore, the production cost for rainbow trout is high as the artificial feed contains high quantity of protein diet (Rai *et al.*, 2008) [25].

The protein component of rainbow trout feed is not only the most important dietary nutrient but also the single most expensive portion. The protein should be biologically available for the rainbow trout in the form of feed and chemically to convert them in the required form (Bekibele *et al.*, 2013) [8]. It provides major basis for growth, development, and reproduction (Steffens, 1989) [31] and (Kaushik, 1995) [19]. In between animal and plant proteins, animal protein is the main dietary component used in formulated diet of rainbow trout, since its introduction in Nepal (Roy *et al.*, 1999) [28] because it contains all essential amino acids especially leucine, isoleucine, methionine, and tryptophan. Because

animal protein is the most costly item which when decreases in cost will sustain rainbow trout cultivation at farmer's level. Fishmeal (FML) which is highly costly is one of the main animal protein supplements in the rainbow trout feed in Nepal due to its high nutrient density (20-35% protein) and digestibility (Nepal *et al.*, 2002) [22] as it contains high level of protein and appreciable quantities of fat and minerals. Therefore, the protein contained in FML has high biological value because of its richness in essential amino acids especially lysine and sulphur-containing methionine and cysteine. Dried trash fish (DTF) which is costly is another main source of animal protein but its poor milling quality and bad smell limits its use (Roy *et al.*, 1999) [28]. Shrimp meal (SML) which is also costly is still another main protein supplement of artificial feed for rainbow trout culture in Nepal (Nepal *et al.*, 2002) [22]. Even though highly costly, FML and SML have become the animal protein supplement for rainbow trout feed in Nepal. Hence, FML and SML increase production cost in rainbow trout cultivation (Anonymous, 2004 [2], Roy *et al.*, 1999 [28]; and Pradhan, 1999 [23]). The dietary protein requirement for rainbow trout ranges from 30-35%. Therefore, most of the studies conducted in Nepal have been focused on finding alternate source of protein supplements in artificial feed of rainbow trout which could be locally available at relatively cheaper price without affecting survival, growth, production, and quality.

**Table 1:** Proximate analysis of feed ingredients of the formulated and control diets

Ingredients	Crude proteins (%)	Crude fibres (%)	Crude fats (%)	Ash (%)	Moisture (%)	Remarks
Silkworm pupae	57.21	2.39	31.29	4.01	6.65	Treatment-1
Silkworm moths	54.55	4.84	29.05	4.74	11.84	Treatment-2
Lysine	100.00					Treatment-3
Methionine	100.00					Treatment-3
Shrimp meals	64.80	3.25	3.81	10.09	10.59	Treatment-4
Soybean (roasted)	37.00	6.00	21.00	5.10	8.70	All the Treatments
Wheat	12.90	9.90	3.80	7.00	12.40	All the Treatments

Very limited numbers of artificial feed ingredients are available to choose for the formulation of fish diet. A review of literature on selected research and investigations into the use of plant feedstuffs in rainbow trout artificial feed showed that it was possible to utilize processed soybean meal at high level (up to 60%) without impairing survival, growth, and environment (Bista *et al.*, 2008) [10]. The mixture of different levels of defatted soybean meal, corn gluten meal, and meat meal could replace up to 90% of the FML, if combination of these ingredients produce the same profile of amino acids comparable to FML (Juadee and Watanabe, 1993) [18]. Grain and byproducts are insufficient as these can't fulfill whole requirement of rainbow trout feed. One of the promising alternatives to the FML and SML is silkworm pupae (SWP), a waste product of silk industry.

SWP, although low cost ingredient, has more protein and lipid than SML (Bhuiyan *et al.*, 1989) [9] and is rich in amino acid profile than FML (Solomon and Yusufu, 2005) [30]. SWP as diet for fingerlings of common carp and Indian major carp has

proven its suitability as substitute of oil cake and rice bran (Chakrabarty *et al.*, 1973) [13] so it will be better substitute of SML. Common carp fed with increasing level of SWP revealed progressive growth along with highest growth in 30% SWP (Cheng *et al.*, 2003) in comparison to diet containing 30% FML (Nandeeshia *et al.*, 1990) [21]. So, SWP could be used as a cheapest and top class unconventional protein and energy artificial feed for rainbow trout after proper processing at reasonable cost. Silkworm moth (SWM), *Bombyx mori* which die after spawning could also be used as another cheapest unconventional protein and energy feed for rainbow trout after proper processing at reasonable cost. Synthetic amino acids (SAA) could further be supplemented in rainbow trout diet as an alternative and unconventional protein feed in comparison to that of animal based protein source. Therefore, artificial feed was formulated using low cost, alternative and unconventional animal protein ingredients of SWP and SWM, and unconventional synthetic protein ingredients of SAA.

**Table 2:** Composition of feed ingredients (%) in the formulated and control diets (Pearson's square method)

Ingredients	SWP	SWM	SAA	SML/control	Remarks
Silkworm pupae	55				Treatment-1
Silkworm moths		60			Treatment-2
Lysine			21		Treatment-3
Methionine			7		Treatment-3
Shrimp meals				50	Treatment-4

Soybean (roasted)	30	30	30	25	Roasted/powder
Wheat	13	8	40	23	All the Treatments
Mineral premixes	1	1	1	1	All the Treatments
Vitamin premixes	0.99	0.99	0.99	0.99	All the Treatments
Vitamin-C premixes	0.01	0.01	0.01	0.01	All the Treatments
Total	100.00	100.00	100.00	100.00	-

## 2. Materials and Methods

The study was conducted for one year from June, 2011 to May, 2012 in farmer's raceways at Kakani, Nuwakot district, Kathmandu, Nepal situated at latitude 27° 48' N, longitude 85° 15' E and altitude 1550 msl. The raceways were supplied with crystal-clear and cold freshwater from permanent, perennial, and dependable water resource (WR) of spring-fed torrential stream.

### 2.1 Physico-chemical parameters

Physico-chemical parameters like water temperature (WT), power of hydrogen ion concentration (pH), dissolved oxygen (DO), free carbon dioxide (FCO), and water discharge (WD) were measured during the research period of 150 days (5 months) from December 2011 to May 2012 following APHA (2005) with the help of a calibrated thermometer (0-100 °C), battery-operated pH meter, standard Winkler's method, Nessler's method, and fill in the bucket method  $\{WD = 100L \div t \text{ (sec)} L \text{ sec}^{-1}\}$  respectively at Water Research Laboratory, KUKL, Kirtipur, Kathmandu, Nepal.

### 2.2 Feed formulations, processing and preparation

Artificial feed was formulated using low cost, alternative, and unconventional animal protein ingredients of SWP

(Treatment-1) and SWM (Treatment-2), and synthetic protein of SAA (Treatment-3) having lysine and methionine (3:1) and then compared to that of SML (Treatment-4) acting as control. Other ingredients taken were locally available soybean and wheat. Mineral and vitamin premixes were mixed as additives.

The three formulated diets of SWP, SWM, and SAA were fed to free swimming fries (FSFs), fries, and fingerlings during exogenous feeding period and evaluated against the diet containing shrimp meals (SML) acting as control through feed efficiency indicators. Compositions (Pearson's square method) of the test formulations are given in Table-2. Calculations of nutrition percentage of the formulated diets based on above composition (Table-2) was also mentioned (Table-3). Proximate analysis of each ingredients (Table-1) and formulated diets (Table-4) were done with reference to crude protein, crude lipid, crude fibre, ash and moisture by Kjeldahl protein analysis method, Soxhlet extraction method, organic residue left method, Moful furnace method, and loss in weight method respectively at Food Research Laboratory, Kathmandu, Nepal and Aquaculture and Limnology Research Unit, Department of Zoology, University of North Bengal, Siliguri, West Bengal, India.

**Table 3:** Calculation of nutrition percentage of the formulated and control diets based on above composition as shown in Table-2

Diets	Crude proteins (%)	Crude fibres (%)	Crude fats (%)	Ash (%)	Moisture (%)	Remarks
SWP/ Formulated	45.25	4.41	24.00	4.79	6.88	Treatment-1
SWM/ Formulated	44.86	5.49	24.30	4.93	10.70	Treatment-2
SAA/ Formulated	44.26	3.96	7.82	4.33	7.57	Treatment-3
SML/Control	44.62	5.41	8.03	7.57	10.33	Treatment-4

Feed processing (drying, heating, grinding, sieving, and screening) of the protein supplements like animal proteins of SWP, SWM, and SML and plant protein of soybean, and energy supplements of wheat were done properly. Feed preparation (mixing, agglomerating, drying, heating, screening, crumbling and pelleting) first taking SWP, second SWM, third SAA, and fourth SML along with soybean, wheat, and additives like mineral premixes (Technovit M), vitamin premixes (Technovit M), and vitamin-C premixes (Technovit C) was done in an efficient manner. The dried ingredients were sieved through a 60 mesh screen and homogenized by blending thoroughly in a feed mixer. The proper amount of water was added to the ingredients to make them dough allowing them to make crumble feed of the size of 180-500  $\mu$  for FSFs and of the size of 600-1000  $\mu$  for fries and pellet feed of the size of 1400-1700  $\mu$  for fingerlings. After crumbling and pelleting, all the four types (3 formulated and 1 control) of artificial feed was air dried and then kept in 50 °C in a constant temperature oven until they reached 8-12% moisture, and then bagged and stored in deep freezer until used.

### 2.3 Experimental protocol

For getting free swimming fries (FSFs) for the research experiment, 1<sup>+</sup>broods were stocked in June 2011, artificial

breeding was done in November 2011, and then, the FSFs were obtained in December 2011. FSFs of rainbow trout with body weight  $0.0406 \pm 0.0018$  g and length  $1.92 \pm 0.097$  were stocked at the density of 250 m<sup>-2</sup> for each feed formulation in quadruplicate nursing cum feeding cum rearing cages (1.0 m  $\times$  1.0 m  $\times$  1.0 m) placed in raceway. In this way, 4000 FSFs (1000 in each set and 250 in each replica) were stocked. Physico-chemical parameters like water temperature (WT), pH, dissolved oxygen (DO), free carbon dioxide (FCO), and water discharge (WD) of the raceways accommodating cages were recorded month-wise during the research period of 150 days (5 months) from December 19, 2011 to May 16, 2012 but WD which was always more than it was required was maintained as per Basnet *et al.* 2008 [7] and Rai *et al.* 2008 [25]. FSFs were fed 45% crude protein (CP) containing crumble and pellet feed according to Basnet *et al.* 2008 [7], Bista *et al.* 2008 [10] and Rai *et al.* 2008 [25]. To do this, they were provided respective feed up to the satiation at the interval of 1 hour for 12 times during day time @ 15% of their live body weight for 30 days (1 month) in WD of 0.083 L sec<sup>-1</sup> m<sup>-2</sup>. FSFs which were grown into fries were supplied respective feed up to the satiation at the interval of 1.2 hours for 10 times during day time @ 12% of their live body weight for 60 days (2 months) in WD of 0.083 L sec<sup>-1</sup> m<sup>-2</sup>. Fries which were grown into fingerlings were given respective feed up to the

satiation at the interval of 1.5 hours for 8 times during day time @ 10% of their live body weight for 60 days (2 months) in WD of 0.083 L sec<sup>-1</sup> m<sup>-2</sup>. Feed cost (of kg<sup>-1</sup> feed) (in NRs) of the formulated and control diets were calculated based on retail and wholesale price of the feed ingredients.

## 2.4 Survival and growth studies

Data of total feed intake (TFI), survival (SR) and growth (GR) (both length and weight) were obtained at every 15 days (two weeks) interval up to 5 months by taking samples of 10 for each (Ricker, 1975). Mortality (MR), SR, TFI, TPI, length (L), GR referring to the weight (W), FE, PER, AGR, SGR, RGR, CF, FCR, PPV, HGP, feed cost (FC) (kg<sup>-1</sup> feed) (NRs), production cost (PC) (kg<sup>-1</sup> trout production) (NRs), and cost analysis (CA) (comparison of FC and PC) was done according to the following formulae given below. Survival and growth including FI, TPI, L, GR (W), FE, PER, AGR, SGR, RGR, CF, FCR, and PPV were calculated using the formulae as follows as:

1. MR (%) = {number of dead fish ÷ number of stocked fish} × 100
2. SR (%) = {number of survived fish ÷ number of stocked fish} × 100
3. TFI (g fish<sup>-1</sup>) = Total feed consumed (g) ÷ number of fish
4. TPI (g fish<sup>-1</sup>) = {FI (g) × CP in the diet (g)} ÷ 100
5. GR in L (cm) = final length (L<sub>2</sub>) – initial length (L<sub>1</sub>)
6. GR in W (g) = final weight (W<sub>2</sub>) – initial weight (W<sub>1</sub>)
7. FE (%) = [{final wt. W<sub>2</sub> (g) – initial wt. W<sub>1</sub> (g)} ÷ mass (dry) of feed consumed] × 100
8. PER (numerical) = {final weight (W<sub>2</sub>) – initial weight

(W<sub>1</sub>) in g} ÷ total protein intake (g)

9. AGR (g day<sup>-1</sup> and g month<sup>-1</sup>) = {final wt. W<sub>2</sub> (g) – initial wt. W<sub>1</sub> (g)} ÷ time t between weightings
10. SGR (% day<sup>-1</sup> and % month<sup>-1</sup>) = [{final wt. W<sub>2</sub> (g) – initial wt. W<sub>1</sub> (g)} ÷ time t between weightings] × 100
11. RGR (%) = [{final wt. W<sub>2</sub> (g) – initial wt. W<sub>1</sub> (g)} ÷ final wt. W<sub>2</sub> (g)] × 100
12. CF (%) = {final wt. W<sub>2</sub> (g) ÷ total length L<sup>3</sup> (cm)} × 100
13. FCR (numerical) = mass (dry) of food consumed ÷ increase in mass (wet) of animal product
14. PPV = total protein intake (g) ÷ feed intake (g) × 100
15. HGP (months) = comparison of growth (L in cm and GR in g) of different months
16. FC (kg<sup>-1</sup> feed) (NRs) = cost of all ingredients making 1 kg of feed
17. PC (kg<sup>-1</sup> trout production) (NRs) = FCR × FC (kg<sup>-1</sup> feed)
18. CA = comparison of PC (kg<sup>-1</sup> trout production cost
19. with that of FC (kg<sup>-1</sup> feed)

## 2.5 Statistical analysis

Data of survival and growth including TFI, TPI, PER, GR (L), GR (W), AGR, SGR, RGR, FE, CF, FCR, and PPV were expressed as mean ± SE and subjected to Anova: Single Factor, Two Factor without Replication and Two Factor with Replication in MS-EXCEL for interactions among Feed Formulations (Treatments). Differences between the two highest and higher data obtained in the results were separated with the help of t-test and F-test to show the significance statistically.

**Table 4:** Proximate analysis of the prepared formulated and control diets

Feed nutrients and others	SWP	SWM	SAA	SML/control	Remarks
Crude proteins (%)	42.68	46.01	48.47	48.27	Crumble/Pellet
Crude fibres	2.80	3.11	1.86	2.65	Crumble/Pellet
Crude lipids	17.01	20.85	6.48	8.62	Crumble/Pellet
Ash	5.65	7.13	5.06	11.67	Crumble/Pellet
Moisture	10.35	3.66	11.54	8.97	Crumble/Pellet

## 3. Results

The present paper describes the preliminary findings of the effect of SWP diet, SWM diet, and SAA diet available in Nepal in the formulated diets of rainbow trout on survival and growth during exogenous feeding and evaluating the formulated diets against SML diet acting as control through total feed intake (TFI) and total protein intake (TPI) along with feed efficiency indicators of feed efficiency (FE), protein efficiency ratio (PER), absolute growth rate (AGR), specific growth rate (SGR), relative growth rate (RGR), condition factor (CF), feed conversion ratio (FCR), and protein productive value (PPV) further including highest growth period (HGP) and cost analyses (CA) as shown in Table-6.

Water temperature from December 2011 to May 2012 ranged 8.4-18.8 (13.46 ± 1.97 °C), pH 7.2-7.9 (7.56 ± 0.12), DO 7.5-10.5 (9.16 ± 0.59 mg L<sup>-1</sup>), free carbon dioxide 1.5-4.5 (3.08 ± 0.60 mg L<sup>-1</sup>) and water discharge was 45-61 (53 ± 2.71 L sec<sup>-1</sup>). Physico-chemical parameters were significant (*P*<0.01). pH and DO were negatively correlated with water temperature, free carbon dioxide and water discharge (Table-5).

FSFs, fries, and fingerlings of rainbow trout were fed three different feed formulations of SWP diet, SWM diet, and SAA diet along with a control diet of SML in which TFI tended to

be 59.054 ± 0.019 g fish<sup>-1</sup>, 10.445 ± 0.842 g fish<sup>-1</sup>, 4.591 ± 0.236 g fish<sup>-1</sup>, and 25.734 ± 0.143g fish<sup>-1</sup> respectively. When calculated, TPI came to be 25.204 ± 0.008 g fish<sup>-1</sup>, 4.806 ± 0.04 g fish<sup>-1</sup>, 2.226 ± 0.11 g fish<sup>-1</sup>, and 12.422 ± 0.07 g fish<sup>-1</sup> respectively with SWP, SWM, SAA, and SML diets. Therefore, SR and GR due to SWP diet in terms of TFI and TPI were significant (*P*<0.01) with that of SWM, SAA and SML diets.

Fingerlings obtained due to SWP, SWM, SAA, and SML diets were 912, 817, 263, and 942 in number respectively. Survival (SR) of the above mentioned stages revealed SML diet fed stages to have 94.20 ± 0.675% during 150 days (5 months) of nursing, feeding and rearing being significantly superior (*P*<0.01) among all the formulated diets. SR of SML diet was comparable to SWP diet with 91.20 ± 1.026% and SWM diet with 81.70 ± 2.107%. However, stages fed with SAA diet had least SR of 26.30 ± 9.327% among all the feed treatments (Table-6). In this way, SR due to feed, SR due to days and feed, and SR due to months and feed were significant (*P*<0.01). However, SR due to SML diet was insignificant with that of SWP diet but SR due to SML diet was significant (*P*<0.01) with that of SWM and SAA diets.

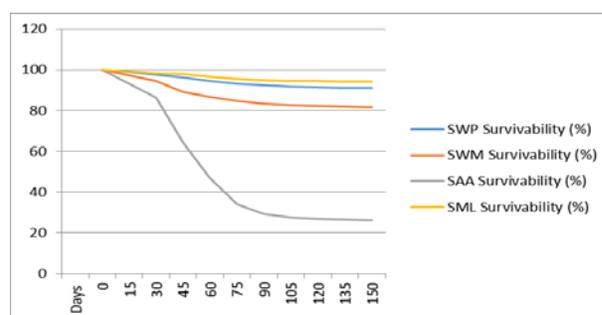
**Table 5:** Physico-chemical parameters of the raceways accommodating cages

S. No.	Months	WT (°C)	pH (1-14)	DO (mg L <sup>-1</sup> )	FCO (mg L <sup>-1</sup> )	WD (L sec <sup>-1</sup> )	Remarks
1.	Jan	8.4	7.9	10.5	1.5	51	Dec 19, 2011 to Jan 18, 2012
2.	Feb	9.9	7.7	10.0	2.2	53	Jan 19, 2011 to Feb 18, 2012
3.	Mar	13.4	7.6	9.8	2.8	45	Feb 19, 2011 to Mar 18, 2012
4.	Apr	16.8	7.4	8.0	4.4	55	Mar 19, 2011 to Apr 18, 2012
5.	May	18.8	7.2	7.5	4.5	61	Apr 19, 2011 to May 16, 2012

Fingerlings obtained due to SWP, SWM, SAA, and SML diets were  $13.36 \pm 0.145$  cm and  $31.278 \pm 0.194$  g,  $6.77 \pm 0.168$  cm and  $3.195 \pm 0.057$  g,  $5.07 \pm 0.214$  cm and  $1.045 \pm 0.092$  g, and  $10.61 \pm 0.194$  cm and  $10.962 \pm 0.194$  g with the growth (GR) of  $11.44 \pm 0.145$  cm and  $31.237 \pm 0.194$  g,  $4.90 \pm 0.214$  cm and  $3.154 \pm 0.057$ g,  $3.15 \pm 0.214$  cm and  $1.004 \pm 0.092$  g, and  $8.69 \pm 0.076$  cm and  $10.921 \pm 0.194$  g respectively. FSFs, fries, and fingerlings grew exponentially with the three feed formulations containing animal protein of SWP, SWM, and SML. The periodic growth of above mentioned stages was somewhat stagnant with feed comprised of SAA (Figure-2). Therefore, GR with SWP diet fed stages exhibited superiority ( $P < 0.01$ ) over other formulated diets against all feed efficiency indicators and that of SAA diet fed stages lowest whereas SML and SWM diets fed stages lied in between. In this way, GR due to feed, GR due to days and feed, and GR due to months and feed significant ( $P < 0.01$ ). Further, GR due to SWP diet was significant ( $P < 0.01$ ) with that of SML, SWM and SAA diets.

FE was highest due to SWP diet ( $52.898 \pm 0.312\%$ ) and lowest due to SAA diet ( $21.753 \pm 1.066\%$ ) and with SML diet ( $42.434 \pm 0.0317\%$ ) and SWM diet ( $30.2 \pm 0.0351\%$ ) in between. Further, PER was highest due to SWP diet ( $1.239 \pm 0.007$ ) and was significantly different ( $P < 0.01$ ) from SML diet ( $0.879 \pm 0.012$ ) which was higher, however, PER was low due to SWM diet ( $0.656 \pm 0.008$ ), and lowest due to SAA diet ( $0.449 \pm 0.022$ ). The highest AGR was obtained with SWP diet fed stages ( $0.208 \pm 0.0013$  g day<sup>-1</sup> and  $6.248 \pm 0.039$  g month<sup>-1</sup>) followed by SML diet fed stages ( $0.0713 \pm 0.0019$  g day<sup>-1</sup> and  $2.134 \pm 0.055$  g month<sup>-1</sup>) and SWM diet fed stages ( $0.021 \pm 0.0004$  g day<sup>-1</sup> and  $0.631 \pm 0.012$  g month<sup>-1</sup>). The lowest AGR was obtained with SAA diet fed stages ( $0.0068 \pm 0.0008$  g day<sup>-1</sup> and  $0.201 \pm 0.019$  g month<sup>-1</sup>). The highest SGR was due to SWP diet ( $20.725 \pm 0.086\%$  daily-wise and  $624.75 \pm 3.886\%$  monthly-wise) and lowest due to SAA diet ( $0.675 \pm 0.075\%$  daily-wise and  $20.05 \pm 1.851\%$  monthly-wise) and with SML diet ( $7.125 \pm 0.189\%$  daily-wise and  $213.43 \pm 5.490\%$  monthly-wise) and SWM diet ( $2.100 \pm 0.041\%$  daily-wise and  $63.05 \pm 1.165\%$  monthly-wise) in between. The RGR was highest due to SWP diet ( $5.129 \pm 0.031\%$  daily-wise and  $153.88 \pm 0.0956\%$  monthly-wise), higher due to SML diet ( $1.752 \pm 0.045\%$  daily-wise and  $52.567 \pm 1.349\%$  monthly-wise), low due to SWM ( $0.518 \pm 0.009\%$  daily-wise and  $15.537 \pm 0.282\%$  monthly-wise), and lowest due to SAA diet ( $0.165 \pm 0.015\%$  daily-wise and  $4.946 \pm 0.454\%$  monthly-wise). There was significant difference ( $P < 0.01$ ) among formulated diets on the growth (GR) of above mentioned stages against all feed efficiency indicators. FE, PER, AGR, SGR, RGR, and were highest due to SWP diet, lowest due to SAA diet while due to SML diet and SWM diet in between. Again, CF was highest due to SWP diet ( $1.89 \pm 0.19$ ), higher due to SML diet ( $1.35 \pm 0.126$ ), low due to SAA diet ( $1.25 \pm 0.08$ ), and lowest due to SWM diet ( $1.23 \pm 0.07$ ). In this way, day-wise and month-wise FE, PER, AGR, SGR, RGR, and CF were significant ( $P < 0.01$ ). Further, FE, PER, AGR, SGR, RGR, and CF due to SWP diet were

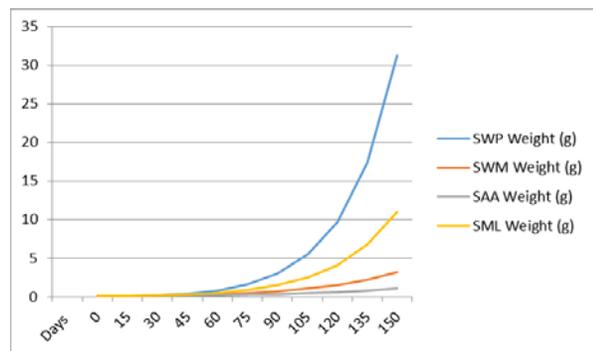
significant ( $P < 0.01$ ) with that of SML, SWM, and SAA diets. Unlike other feed indicators, FCR with SAA diet fed stages exhibited highest conversion rate of  $4.631 \pm 0.228$  being significantly different ( $P < 0.01$ ) with that of SWP diet being lowest conversion rate of  $1.897 \pm 0.0074$ , however, conversion rate of  $2.358 \pm 0.031$  and  $3.313 \pm 0.0376$  respectively with SML diet and SWM diet were intermediate. PPV was highest due to SAA diet ( $48.48 \pm 0.002\%$ ), higher due to SML diet ( $48.27 \pm 0.0002\%$ ), low due to SWM diet ( $46.01 \pm 0.002\%$ ), and lowest due to SWP diet ( $42.68 \pm 0.00\%$ ) (Table-6). Hence, FCR and PPV were significant ( $P < 0.01$ ). Further, FCR due to SWP diet was significant ( $P < 0.01$ ) with that of SML, SWM, and SAA diets.

**Fig 1:** Survivability of FSFs, fries, and fingerlings in 150 days (5 months)

The HGP was observed during April 20 to May 19, 2012 and lowest in the period of December 21, 2011 to January 15, 2012 whereas it was medium in the period of March 21 to April 4, 2012. Cost analyses (production cost of rainbow trout and cost of feed) revealed that SWP diet kg<sup>-1</sup> was NRs 73.45 with production cost of rainbow trout kg<sup>-1</sup> NRs 139.30, SWM diet NRs 49.45 with production cost NRs 163.80, SAA diet NRs 167.95 with production cost NRs 777.75, and SML diet NRs 193.45 with production cost NRs 456.15.

#### 4. Discussions

Rainbow trout mostly requires glacier water or clean cold spring water for its successful breeding, survival and growth (Basnet *et al.* 2008)<sup>[7]</sup>. Water resource (WR) supplying water in the raceways, in the present work, was permanent, dependable, and from perennial spring-fed stream. The prerequisite for rainbow trout culture is adequate volume of coldwater below 20 °C because feed consumption decreases when WT increases above 20 °C, resulting into slow growth and eventually death, if exposed to higher (always more than 20 °C) WT for a longer period (Rai *et al.* 2008)<sup>[25]</sup>. What so ever may be, rainbow trout require WT 0-25 °C (Swar, 2008)<sup>[32]</sup> but according to Yamazaki (1991)<sup>[33]</sup>, it grows well in WT 10-18 °C, however, according to Anonymous (2001)<sup>[5]</sup>, its best growth in Nepal occurs in WT 16-18 °C. WT in the present work was 8.4-18.8 °C. When WT increased then FCO and WD increased but pH and DO decreased but when WT decreased vice-versa. Hence, FCO and WD positively correlated with WT and pH and DO negatively.



**Fig 2:** Growth of FSFs, fries, and fingerlings in 150 days (5 months)

The preferred range of pH for rainbow trout is 6.5-8.0 with optimum value 7.0-7.5 (Rai *et al.*, 2008) [25] for semi-intensive farming because at higher pH levels, relatively low levels ammonia (NH<sub>3</sub>) can be dangerously toxic (Bromage and Shephard, 1990) [12] and (Sedgwick, 1985) [29]. The pH in the present investigation was 7.2-7.9. According to Huet (1975) [17], rainbow trout require DO above 7.0 mg L<sup>-1</sup>; according to Rai *et al.* (2008) [25], it requires DO more than 7.0 mg L<sup>-1</sup>; and according to Basnet *et al.*, (2008) [7], its brood requires cold, clean and high DO containing water of 7.0-7.5 mg L<sup>-1</sup> for normal trout culture and 10-11 mg L<sup>-1</sup> for intensive culture for proper ripening of gonads and successful hatching of alevins and FSFs because according to Anonymous (1998) [4] the growth is retarded and the trout may die, if exposed to DO below 7.0 mg L<sup>-1</sup>. In the present research, DO ranged in between 7.5-10.5 mg L<sup>-1</sup>. Rainbow trout require FCO below 20mg L<sup>-1</sup> (Lawson, 2011) [20]. So, induced breeding under semi-intensive culture was done in the present work because pH was 6.7-7.9 and DO below 11 mg L<sup>-1</sup>. FCO in the present

work was 1.5-4.5 mg L<sup>-1</sup>. Hence, physico-chemical parameters of the research period of 150 days from December 19, 2011 to May 16, 2012 (5 months), in the present investigation, were suitable for the SR and GR of FSFs, fries, and fingerlings, however, WD which was more than it was required was maintained 0.083 L sec<sup>-1</sup> m<sup>-2</sup> as per requirement mentioned above.

Highest TFI of SWP diet, higher of SML diet, low of SWM diet, and lowest of SAA diet was due to the quantity of feed consumed by FSFs, fries, and fingerlings to attend the survival and growth mentioned. Hence, TPI came to be highest due to SWP diet, higher due to SML diet, low due to SWM diet, and lowest due to SAA diet because of TFI. Higher SR and highest GR due to SWP diet in comparison to SML, SWM, and SAA diets was because of the highest TFI along with highest TPI and PER.

Highest SR of the above mentioned stages due to SML fed diet was because of the presence of animal protein (CP 47.27%) with required essential amino acids. Higher SR due to SWP diet was because of the slightly less quantity of animal protein (CP 42.68%). So, SR due to SML diet was near SWP diet.

Highest GR of the above mentioned stages with SWP fed diet was due to the presence of required amount of essential amino acids and fats in the diet. Higher GR of the above mentioned stages with SML fed diet was due to the presence of required amount of essential amino acids but fewer amounts of fats in the diet. So, GR due to SWP diet was far from SML diet.

FE, PER, AGR, SGR, and RGR were highest due to SWP diet, higher due to SML diet, low due to SWM diet, and lowest due to SAA diet totally depending up on TFI and TPI. CF was higher due to the SML diet because of the fewer

**Table 6:** Survival and growth of FSFs, fries, and fingerlings of rainbow trout due to 3 formulated diets (SWP, SWM, and SAA) evaluated with control (SML) diet during the period of 150 days (5 months)

Particulars	SWP	SWM	SAA	SML/CON	Remarks
Animal or synthetic proteins (kg <sup>-1</sup> diet)	0.55	0.60	0.28	0.50	
CP (%)	42.68	46.01	48.47	48.27	
FSFs (number) stocked	1000	1000	1000	1000	
FSFs (cm) stocked	1.92±0.097	1.92±0.097	1.92±0.097	1.92±0.097	
FSFs (g) stocked	0.0406±0.0018	0.0406±0.0018	0.0406±0.0018	0.0406±0.0018	
FI (g fish <sup>-1</sup> )	59.054±0.0192	10.445±0.842	4.591±0.236	25.734±0.143	
TPI (g fish <sup>-1</sup> )	25.204±0.008	4.806±0.04	2.226±0.11	12.422±0.07	
Research period (days)	150	150	150	150	5 months
Fingerlings (number) obtained	912	817	263	942	
Fingerlings (cm) obtained	13.36±0.145	6.77±0.168	5.07±0.214	10.61±0.194	
Fingerlings (g) obtained	31.278±0.194	3.195±0.057	1.045±0.092	10.962±0.194	
Survival (%)	91.20±1.026	81.70±2.107	26.30±9.326	94.20±0.675	
Growth (cm)	11.44±0.145	4.90±0.214	3.15±0.214	8.69±0.076	
Growth (g)	31.237±0.194	3.154±0.057	1.004±0.092	10.921±0.194	
FE (%)	52.898±0.312	30.2±0.351	21.753±1.066	42.434±0.031	
PER (numerical)	1.239±0.007	0.656±0.008	0.449±0.022	0.879±0.012	
AGR (g day <sup>-1</sup> )	0.208±0.0013	0.021±0.0004	0.0068±0.0008	0.0713±0.0019	
AGR (g month <sup>-1</sup> )	6.248±0.039	0.631±0.012	0.201±0.019	2.134±0.055	
SGR (% day <sup>-1</sup> )	20.725±0.086	2.100±0.041	0.675±0.075	7.125±0.189	
SGR (% month <sup>-1</sup> )	624.75±3.886	63.05±1.165	20.05±1.851	213.43±5.490	
RGR (% day <sup>-1</sup> )	5.129±0.031	0.518±0.009	0.165±0.015	1.752±0.045	
RGR (% month <sup>-1</sup> )	153.88±0.956	15.537±0.282	4.946±0.454	52.567±1.349	
CF (%)	1.89±0.19	1.23±0.07	1.25±0.08	1.35±0.126	
FCR (ratio)	1.897±0.0074	3.313±0.0376	4.631±0.228	2.358±0.031	
PPV (%)	42.68±0.00	46.01±0.002	48.48±0.002	48.27±0.0002	
FC (kg <sup>-1</sup> feed)	73.45	49.45	167.95	193.45	NRs
PC (kg <sup>-1</sup> trout production)	139.30	163.80	777.75	456.15	NRs

amounts of fats, and highest due to SWP diet because of the more amount of fats. FCR due to SWP diet was lowest due to the reason of more GR. Further, PPV was highest due to SAA diet, higher due to SML diet, low due to SWM diet, and lowest due to SWP diet because of the percentage of the CP present in them. Therefore, the results of PPV exactly matched the percentage of the CP obtained in the proximate analyses of the formulated and control diets (Table-5).

The HGP observed during April to May was due to required quantity of physico-chemical parameters. CA revealed SWM diet cheapest with low production cost, SWP diet cheaper with lowest production cost, SAA diet cheap with highest production cost, and SML diet costly with high production cost. SWP diet showed lowest production cost because of its remarkable SR and excellent GR.

Rainbow trout fed FML diet containing 37% CP grew as fast as those fed 42% CP diets supplemented with SAA containing lysine, methionine, threonine, and tryptophan (Cheng *et al.*, 2003) [14]. Further, reduction (of 2.7%) of dietary digestible CP from 27.0 to 24.3% with SAA had no negative impact on growth performance of Nile tilapia (Botaro *et al.*, 2007) [11]. Again, dietary crude protein (CP) could be reduced from 41.26 to 35.52% in the diets of *L. vannamei* as long as SAA were supplemented (Huai *et al.*, 2008) [16]. However, SR and GR were poorest due to the diet of SAA which may be due to the absence of animal protein with required amino acid profile and insufficient lipid due to required fatty acid profile. Further, GR due to SWP diet was significant with SWM, SAA and SML diets.

SWP, although low cost ingredient, has more protein and lipid than SML (Bhuiyan *et al.*, 1989) [9] and is rich in amino acid profile than FML (Solomon and Yusufu, 2005) [30]. Hence, feed formulation with SWP diet had exhibited superiority in GR over all other feed formulations against all feed efficiency indicators (Table-6), however less SR than SML diet might be due to more fatty acid in SWP diet than it was required. Because SWP as diet for fingerlings of common carp and Indian major carp has proven its suitability as substitute of oil cake and rice bran (Chakrabarty *et al.*, 1973) [13] so it will be better substitute of SML diet. Because common carp fed with increasing level of SWP revealed progressive growth along with highest growth in 30% SWP (Cheng *et al.*, 2003) in comparison to diet containing 30% FML (Nandeeshha *et al.*, 1990) [21] hence, above mentioned stages of rainbow trout had shown highest growth with SWP in comparison to SML.

## 5. Conclusions

Results indicated that diets with natural and animal protein of SWP, SWM, and SML were superior to diet with synthetic protein of SAA. Results also indicated that survival due to SML diet was slightly more than SWP diet but insignificant. Results further indicated that growth due to SWP diet was highly more than SML diet and highly significant also. Therefore, TFI and TPI of SWP diet was more than others. Again, SWP diet was highly significant in terms of FE, PER, and FCR in comparison to other diets. Further, SWP diet was superior in AGR, SGR, RGR, CF, and CA in comparison to rest of the diets. Hence, the results finally confirmed that cost effective SWP diet could be used as better alternative to completely replaced SML diet without compromising survival and growth of rainbow trout.

## 6. Acknowledgements

The authors would like to thank Mr. Nand Kishor Roy, Senior

Technical Officer (Fish Nutrition) for his continuous cooperation and help in the present study. They would further like to thank Mr. Gyanendra Bahadur Karki, Chief (Deputy Manager) and Mr. Ram Hulas Jha, Laboratory Officer, KUKL, Water Research Laboratory, Kirtipur, Kathmandu, Nepal. They would also like to thank the Laboratory Analysts who helped in the analyses of the artificial feed of the rainbow trout broods. Thanks are also due the fishermen who were directly involved in the present study especially during breeding of the rainbow trout.

## 7. References

1. American Public Health Association (APHA). Standard Methods for the Examination of Water and Waste Water. AWWA, WPEC, 21<sup>st</sup> Edition, Washington DC, USA. 2005.
2. Anonymous. Comparative Study of Growth and Meat Quality of Rainbow Trout Fed with Different Levels of Proteins. Annual Technical Report (2003/2004). Fisheries Research Centre, Trishuli, Nuwakot, Nepal. 2004, 14-20.
3. Anonymous. Annual Technical Report (2002/2003). Nepal Agricultural Research Centre, Fisheries Research Division, Godawari, Lalitpur, Nepal, 2003.
4. Anonymous. Feed Management Guidelines for Salmonids. Gibson's Limited, Tasmania. 1998, 150.
5. Anonymous. Development of starter feed for trout alevins. Annual Technical Report. Nepal Agricultural Research Centre, Fisheries Research Division, Godawari, Lalitpur, Nepal. 2001, 51-54.
6. Bardach JE, Ryther JH, McLarney WO. Aquaculture - The farming and husbandry of freshwater and marine organisms. John Wiley and Sons Inc., NY. 1972, 868.
7. Basnet SR, Lamsal GP, Mulmi RM, Gurung TB. Breeding performance of rainbow trout (*Oncorhynchus mykiss*) in northeastern hills, Nepal. In: Rainbow trout (*Oncorhynchus mykiss*) farming strategies in Nepal. Proceedings of First National Workshop on scaling up of Rainbow Trout (*Oncorhynchus mykiss*) Farming Strategies in Nepal. Gurung TB (ed.). FRD (NARC), JICA, DIFID (DoA) and NEFIS. 2008, 12-16.
8. Bekibele DO, Ansa EJ, Agokei OE, Opara JY, Alozie-Chidi VC, Aranyo AA *et al.* The Comparative Effect of Fish and Blood Meal Based Diets on the Growth and Survival of Juvenile Tilapia (*Oreochromis niloticus*) in Concrete Tank. Journal of Fisheries and Aquatic Science. 2013; 8(1):184-189.
9. Bhuiyan AKMA, Begum NN, Begum M, Hoq ME. Survey of potential fish feed ingredients of Bangladesh on the basis of their availability and biochemical composition. FRI Research Progress Report No. 1. Freshwater station. 1989, 70.
10. Bista JD, Wagle SK, Pradhan N, Roy NK. Nutrition and Pellet Feed Formulation for Rainbow Trout (*Oncorhynchus mykiss*) in Nepal. In: Rainbow trout (*Oncorhynchus mykiss*) farming strategies in Nepal. Proceedings of First National Workshop on scaling up of Rainbow Trout (*Oncorhynchus mykiss*) Farming Strategies in Nepal. Gurung TB (ed.). FRD (NARC), JICA, DIFID (DoA) and NEFIS. 2008, 32-40.
11. Botaro D, Furuya WM, Silva LCR, Santos LDD, Silva TSDC, Santos VGD. Dietary protein reduction based on ideal protein concept for Nile tilapia (*Oreochromis niloticus*) cultured in net pens. Zootecnia. 2007; 36:517-525.

12. Bromage NR, Shephard CJ. Fish, their requirements and site evaluation. In: Shephard CJ and Bromage NR (eds.). Intensive Fish Farming. BSP Professional Books, Oxford. 1990, 17-49.
13. Chakrabarthy RD, Sen PR, Kowtal GV. Observations on the relative usefulness of different feeds for carp spawn and fry. Journal of Inland Fishery Society of India. 1973; 5:182-188.
14. Cheng ZJ, Hardy RW, Usry JL. Plant protein ingredients with lysine supplementation reduce dietary protein level in rainbow trout (*Oncorhynchus mykiss*) diets, and reduce ammonia nitrogen and soluble phosphorus excretion. Aquaculture. 2003; 218:553-565.
15. Hasan MR. Nutrition and Feeding for Sustainable Aquaculture Development in the Third Millennium. In: Aquaculture in the Third Millennium. Technical Proceedings of the Conference and Aquaculture in the Third Millennium. 2001.
16. Huai MY, Liu YJ, Tian LX, Deng SX, Xu AL, Gao W *et al.* Effect of dietary protein reduction with synthetic amino acids supplementation on growth performance, digestibility, and body composition of juvenile Pacific white shrimp, *Litopenaeus vannamei*. Aquaculture. 2010; 244:174-179.
17. Huet M. Breeding and Cultivation of Salmonids, a Fish Culture in Cold Water. In: Textbook of fish culture, breeding and cultivation of fish. Fishing News Books Ltd., 23 Resemount Avenue, West Byfleet, Surrey, England. 1975, 59-112.
18. Juadee P, Watanabe T. Replacement of fishmeal by alternative protein sources in rainbow trout diets. Proceedings on Fisheries, Department of Fisheries, Bangkok, Thailand. 1993, 15-17.
19. Kaushik SJ. Amino acid requirements, protein and energy utilization. In: From Feed to Feed Symposium: Technological and Nutritional Aspects of Safe Feed Production. Utrecht, Netherlands. 1995, 1-9.
20. Lawson EO. Physico-Chemical Parameters and Heavy Metal Contents of Water from the Mangrove Swamps of Lagos Lagoon, Lagos, Nigeria. *Advances in Biological Research*. 2011; 5(1):8-21.
21. Nandeeshha MC, Srikanth GK, Vargheses P, Keshavappa TJ, Basavaraja N *et al.* Effect of non-defatted silkworm pupae in diets on the growth of common carp, *Cyprinus carpio*. *Biological Wastes*. 1990; 33:17-23.
22. Nepal AP, Basnet SR, Lamsal GP, Joshi PL, Mulmi RM. Economics of Rainbow Trout in Nepal. In: Cold Water Fisheries in the Trans Himalayan Countries. Petr T and Swar DB (eds.). FAO Fisheries Technical. 2002; 376:431.
23. Pradhan N. Development of starter feed for trout larvae under local management. In: Proceedings of the Present Status of Fisheries Research, Development and Education in Nepal. Pradhan BR, Wagle SK, Yamada O and Takano M (eds.). 1999, 66-67.
24. Pradhan N, Raymajhi A, Shrestha SK. Rainbow Trout (*Oncorhynchus mykiss*) Larval Rearing Methodologies in Nepal. In: Rainbow trout (*Oncorhynchus mykiss*) farming strategies in Nepal. Proceedings of First National Workshop on scaling up of Rainbow Trout (*Oncorhynchus mykiss*) Farming Strategies in Nepal. Gurung TB (ed.). FRD (NARC), JICA, DIFID (DoA) and NEFIS. 2008, 17-22.
25. Rai AK, Gurung TB, Basnet SR, Mulmi RM. Present Status and Prospect of Rainbow Trout (*Oncorhynchus mykiss*) Farming in Nepal. In: Rainbow trout (*Oncorhynchus mykiss*) farming strategies in Nepal. Proceedings of First National Workshop on scaling up of Rainbow Trout (*Oncorhynchus mykiss*) Farming Strategies in Nepal. Gurung TB (ed.). FRD (NARC), JICA, DIFID (DoA) and NEFIS. 2008, 5-11.
26. Ricker WE. Computation and Interpretation of Biological Statistics of Fish Populations. Bulletin of the Fisheries Research Board of Canada, Bulletin No. 191, Ottawa. 1975.
27. Robinson EH, Li MH. A Practical Guide to Nutrition, Feeds and Feedings of Catfish. MSU Cares, Mississippi, Agriculture and Forestry Experiment Station, Bulletin, 1041. 1996.
28. Roy NK, Nepal AP, Basnyat SR. Variation in the Growth of Rainbow Trout Fed upon Pellet Feed with Different Proportions of Shrimp Meal. In: Proceedings of the Present Status of Fisheries Research, Development and Education in Nepal. Pradhan BR, Wagle SK, Yamada O and Takano M (eds.). 1999, 47-52.
29. Sedgwick SD. Trout Farming Handbook, 4<sup>th</sup> Edition. Fishing News Books, England. 1985, 160.
30. Solomon SG, Yusufu II. The amino acid profile, proximate and mineral composition of silkworm caterpillar (*Anaphe infracta*) meal as possible alternative to fishmeal in the diets of cultured fish species. Department of Fisheries and Aquaculture. University of Agriculture. 2005.
31. Steffens SH. Principles of Fish Nutrition. Ellies Horward, Chichester, UK. 1989, 384.
32. Swar DB. History of Rainbow trout (*Oncorhynchus mykiss*) introduction in Nepal. In: Rainbow trout (*Oncorhynchus mykiss*) farming strategies in Nepal. Proceedings of First National Workshop on scaling up of Rainbow Trout (*Oncorhynchus mykiss*) Farming Strategies in Nepal. Gurung TB (ed.). FRD (NARC), JICA, DIFID (DoA) and NEFIS. 2008, 1-4.
33. Yamazaki T. Culture of Foreign Fishes Farming, Japan. 25<sup>th</sup> Anniversary. 1991; 25(1):41-46.