Effect of dietary digestible energy level on growth indices of kutum (*Rutilus frisii kutum* Kamenskii, 1901)

Noverian H.A.[1*] ; Shabanipour N. [2]; Khoshkholgh M.R.[1]; and Hosseini M.R.[3]

hamidnav@yahoo.com

1-Department of Fisheries, Faculty of Natural Resources, University of Guilan, P.O. Box: 1144, Somehesara, Iran

2- Department of Biology, Faculty of Basic Sciences of Guilan University, P.O. Box: 41335-1914 Rasht, Iran

3-Iranian Fisheries Research Organization, P.O.Box: 14155-6116 Tehran, Iran

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Abstract: A 60-days feeding trail was conducted to evaluate the effects of dietary digestible energy levels (DE) on kutum, *Rutilus frisii kutum*. Four isonitrogenous (35% protein) diets with digestible energy levels of 2500, 2600, 2700 and 2800kcal kg^{-1} were tested. Two hundred and forty advanced fry (2±0.4g) were randomly distributed in 12 fiberglass tanks of 400L capacity. Nutritional responses in terms of WG, FCR and PER as well as survival rate (SR) significantly improved ($P<0.05$) with increase in DE level from 2500 to 2800kcal kg^{-1} diet, but no difference between 2600 and 2700kcal kg^{-1} diet was found. Body crude protein and fat significant were increased ($P<0.05$) when the dietary energy was raised up from 2500 to 2600kcal DE kg^{-1} diet, but a further increase on energy did not improve the fish crude protein and fat content.

Keywors: *Rutilus frisii kutum*, Nutritional evaluation, Digestible energy, Growth performance

* **Corresponding author**

Introduction

Kutum (*Rutilus frisii kutum*), locally known as Mahi-Sefid, is an endemic cyprinid of the southern Caspian Sea that has been adapted to brackish water as well as surrounding freshwater bodies (Razavi Sayad, 1984; Abbasi *et al*., 2001). Various factors such as overfishing, environmental pollution, decreasing average weight of brood stocks and also declining recruitment in recent decade (1990-2000) have contributed to a sharp decline in kutum stocks (Kazeroni, 1995; Khaval, 1999). Hence, the only alternative for enhancing kutum stocks is to produce fingerlings with appropriate weight using formulated feed adopted for its nutritional requirement prior to their release to the sea.

Studies carried out on the early stages of this fish in earthen ponds indicated that this fish, in addition to natural and wet feeds, is able to consume artificial feeds in a confined area (Danesh, 1992; Noverian *et al*., 2004). Although the macro-nutrient requirements like protein of the advanced fry stage have almost been studied (Noverian *et al*., 2004), other nutritional requirements are unknown and studies are much limited. Therefore, in order to optimize starch and lipid as energy-sparing, the appropriate digestible energy has to be determined. The present study was an attempt to form a base for further macro-nutrients interaction research for kutum.

Materials and methods

Four isonitrogenous (35%) diets were formulated to contain energy of 2500, 2600, 2700 and 2800kcal DE kg^{-1}. The proximal composition of natural and purified ingredients used for the experimental diets are shown in Table 1. All diets were formulated using Lindo-program, 1994. The composition of the experimental diets is shown in Table 2.

All the common natural and purified ingredients, prepared from the local market in one batch, were dried, ground, sieved and stored in air-tight containers. The imported egg albumin, which had the maximum protein content (93%) amongst all the ingredients, was used to adjust the protein level in the experimental diets. Dextrin and oils with 95% starch and 98% fat used as purified ingredients to adjust DE. All natural diets were oven-dried and well pulverized and mixed with purified ingredients; finally, they were fortified with vitamins, minerals and other additives.

Table 1: Proximal composition (as fed basis) of the ingredients used for feed

Ingredients	CP (%)	EE (%)	CF (%)	Ash (%)	NFE (%)	Moisture (%)	Impurity (%)	DE (kcal kg^{-1})
Dextrin	0.58	0	1	1.2	95.0	2.2	0	2860
Soya bean meal	41	2.2	4.3	6.7	27.8	14	4	2300
Fish meal	65	3	0.5	12	6.5	11	2	3200
Rice bran	12	3.5	12	4.2	50.3	12	6	1750
Egg Albumin	93	0	0	3.5	1.2	2.3	0	3570
Wheat flour	18	4.1	6.1	3.9	51.4	12	4.5	2050
Sunflower oil	0	98	0	0	0	2	0	7840
Fish oil	0	98	0	0	0	2	0	7840

CP= crude protein (N × 6.25) **CF= crude fibers** **EE= ether extract**

NFE= nitrogen free extract

DE= Digestible energy was calculated based on animal protein 4.25kcal g^{-1}, plant protein 3.8 kcal g^{-1}, carbohydrate of non-legumes 3 kcal g^{-1}, carbohydrate of legumes 2kcal g^{-1} and fat 8kcal g^{-1} (ADCP, 1983).

All the ground and sieved (200µ) were mixed manually, except the wheat flour that was first gelatinized with distilled water carefully and added later. For obtaining stiff dough, the dough was steamed for 10-15 min without pressure and extruded through a kitchen noodle maker with a 1mm diet to obtain strands (pellet size was 1×1mm). The strands were oven-dried at 60°C for 24h in order to reduce moisture to 10% (Lovell, 1998).

The natural and purified ingredients were blended in appropriate quantities (Table 2), in order to obtain 4 isonitrogenous (35%) diets with different levels of DE.

Table 2: Composition of the experimental diets (% as fed basis)

Ingredients	I (%)	II (%)	III (%)	IV (%)
Dextrin	3.9	5.6	7.3	9
Soya bean	15.7	16	16.3	16.7
Fish meal	9.5	10	10.5	11
Rice bran	19.9	19.5	16	13
Egg albumin	18	18	18	18
Wheat flour	19.9	15.7	15.5	14.7
Sunflower oil	1.5	1.9	2.3	2.7
Fish oil	3	3.8	4.6	5.4
Vit [1]	3	3	3	3
Mineral[2]	3	3	3	3
Binder	3.03	3.03	3.03	3.03
Anti-fungi	0.25	0.25	0.25	0.25
Anti-oxidant	0.02	0.02	0.02	0.02
Vit C	0.2	0.2	0.2	0.2
Proximate composition				
CP	35±0.12	35±0.24	35±0.28	35±0.16
EE	6.5±0.03	7.65±0.56	8.63±0.1	9.69±0.21
CF	4.49±0.23	4.04±0.13	3.69±0.17	3.32±0.11
Ash	4.45±0.18	4.4±0.18	4.35±0.12	4.30±0.13
NFE	28.65±0.28	28.51±0.27	28.38±0.22	28.22±0.99
Moisture	8.5±0.26	8.22±0.19	7.93±0.21	7.65±0.27
AIA[3]	2.91	2.72	2.52	2.33
DE (kcal kg^{-1})	2500±10	2600±12	2700±7	2800±4
E/P (kcal energy g $protein^{-1}$)	71.43	79.29	77.14	80

1: Vitamin mixture (Kanazawa, 1984)
2: U.S.P. mixture XIV from m/s Sisco Research Laboratories
3: A.I.A (Acid Insoluble Ash)

The kutum advanced fries (2±0.4g) were supplied by Shahid Ansari Hatchery and brought to the hatchery facilities at Faculty of Natural Resources (University of Guilan, Iran) and acclimatized in $5m^3$ fiberglass tanks for 4h without feeding prior to the experiment.

The acclimatized kutum fries were carefully weighed with an electronic balance and then transferred randomly into 12 fiberglass tanks of 400L capacity that were filled with 300L filtered freshwater; each tank contained 20 advanced fry (2±0.4g) with continuous aeration. All the experimental trials were conducted in triplicates. The water quality parameters in experimental tanks were monitored daily (Table 3).

Table 3: Mean parameters in experimental tanks

	Months of Rearing	
	June	**July**
Temperature (°C)	25±1.6	27±2.2
pH	7.4±0.31	7.7±0.48
DO_2 (mg/l)	5.3±0.44	6.2±0.67
Total hardness	131±32	126±14

Pelleted diet was provided at the rate of 6% of the body weight 3 times daily (08:00, 12:00 and 16:00h). Excess feed and faeces were removed through siphoning and the culture water was top-upped, each morning.

The experiment was terminated on day of 60 and final weights were recorded and the fish carcass was analyzed for crude protein, lipid (EE), ash, crude fiber and NFE. All ingredients, feed and fish samples were analyzed according to (A.O.A.C, 1985) at Nutritional Lab of the Natural Resources Faculty (Guilan University) and Des were calculated according to ADCP (1983).

The following parameters were estimated:

1. Initial (Wi) and final (Wt) weights to the nearest of 0.1g were recorded.
2. Absolute growth or weight gain (WG) was calculated following Hopkins (1992).
3. Food Conversion Ratio (FCR)= Dry feed consumed (g) / wet weight gain (g)
4. Protein Efficiency Ratio (PER)= Weight gain (g) / protein consumed (g)

5. Survival rate = Final number of fish / Initial number × 100

Statistical analysis of the raw data was done by ANOVA (one way), using SAS and Excel programs. Duncan test was used to determine the significance (95%) of the mean differences.

Results

In this study, kutum fry readily accepted all the experimental diets. The average survival rate (SR) and weight gain significantly improved ($P<0.05$) by increasing digestible energy (DE) in an isonitrogenous diet from 2500 to 2800kcal/kg (Table 4). In addition, the diet performance (FCR and PER) also increased ($P<0.05$) when the dietary energy was increased. When testing the mean growth factors within treatments, no significant differences ($P>0.05$) in all growth indices between treatments 2600 and 2700kcal kg^{-1} were observed. Treatment 2500kcal kg^{-1} with the least DE showed significant difference with the other treatments ($P<0.05$).

Table 4: Preformance of *Rutilus frisii kutum* advanced fry fed experimental diets (DEs) for 60 days (x±SD)

DE (kcal kg)	Initial weight (g)	Final weight (g)	Weight gain (g)	Survival rate (%)	FCR	PER
2500	2.10±0.23[a]	3.88±0.33[a]	1.78±0.21[a]	56±2[a]	2.53±0.27[a]	0.21±0.011[a]
2600	1.98±0.31[a]	4.43±0.29[b]	2.45±0.28[b]	76±3[b]	1.71±0.19[b]	0.75±0.014[b]
2700	2.0±0.34[a]	4.46±0.31[b]	2.46±0.34[b]	77±1[b]	1.73±0.12[b]	0.78±0.019[b]
2800	2.2±0.38[a]	5.69±0.28[c]	3.49±0.32[c]	88±2[c]	1.33±0.10[c]	1.31±0.023[c]

Similar letters within a column imply no significant difference (P>0.05).

Fish crude protein and fat content increased and ash content decreased significantly (P<0.05) when the DE level in the diet was increased from 2500 to 2600kcal DE kg^{-1} (Table 5), but with further increment in DE level, none of the aforementioned contents was significantly affected (P>0.05). Interestingly, the fish carbohydrate content was not significantly affected (P>0.05) by the changes in DE levels.

Table 5: Body composition of advanced fry (on dry matter basis) fed with four levels of DE

DE (kcal kg^{-1})	CP (%)	EE (%)	CF (%)	Ash (%)	NFE (%)
2500	58.1 ± 0.12^a	10.1 ± 0.1^a	3.6 ± 0.22^a	6.2 ± 0.31^a	22.0 ± 0.81^a
2600	60.1 ± 0.17^b	10.6 ± 0.21^b	3.2 ± 0.30^a	3.2 ± 0.18^b	22.9 ± 0.72^a
2700	61.1 ± 0.14^b	10.7 ± 0.11^b	3.0 ± 0.19^a	2.1 ± 0.34^b	23.1 ± 0.91^a
2800	61.5 ± 0.19^b	11.9 ± 0.16^b	2.6 ± 0.42^a	2.0 ± 0.71^b	23.0 ± 0.71^a

Similar letters within a columns imply no significant difference (P>0.05).

Discussion

There is little information available on the macronutrient requirements in terms of energy yielding-nutrients such as starch and lipid (energy sparing) in kutum fry. Since kutum belongs to Cyprinidae family and it has a feeding habit almost similar to common carp (Astramova, 1979; Eross, 1982), the available data on common carp are usually used in the feed formulation of kutum. The designated protein level (35%) in this experiment was based on the findings of an earlier study (Noverian *et al.*, 2004).

Results of the present study on kutum indicated that when the DE in a 35% protein diet was increased from 2500 to 2800kcal kg^{-1}, the survival rate (SR), weight gain and diet performance (FCR and PER) improved (Table 4), which corresponds with the results observed on common carp weighing 8g (Astramova, 1978). Since the optimal dietary energy level could not be determined in this study, further research is required.

Lengyel *et al.* (2001) and Cui & Wootton (1988) recommended the use of starch as an energy source at levels more than 25% in Cyprinidae. This is comparable to present research in kutum when starch inclusion up to 28% was made (Table 2).

Lipid is speculated to play a more important role as an energy source in carnivorous fish like rainbow trout (Garcia-Riera, 1993; Cui & Wootton, 1988) in which the availability of dietary carbohydrates for energy are limited (Cho *et al.*, 1982).

Kutum body protein and fat improved showed when the dietary energy was increased from 2500 to 2800kcal kg^{-1} (Table 5), which indicated the normal distribution of energy, i.e., sparing action of lipid and carbohydrate to protein in metabolism pathway. Similar results have been reported for advanced fry of common carp weighing more than 2g in size (Lengyel *et al.*, 2001; Eross, 1982; Oberie *et al.*, 1997).

Other published reports on kutum are not available for comparison. Hence we hope these results provide some basis for future nutritional research on *Rutilus frisii kutum.*

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