

## UTILIZATION OF AMINO ACIDS IN NILE TILAPIA (*OREOCHROMIS NILOTICUS*) FRY.

### 2-RESPONSE OF NILE TILAPIA FRY TO DIETARY ESSENTIAL AMINO ACIDS DEFICIENCY.

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#### SUMMARY

Twelve semi-purified diets were formulated to detect the influence of 10 dietary essential amino acid deficiency on Nile tilapia, (*Oreochromis niloticus*) fry performance. The first diet was formulated for high control (30% CP) and the last diet (12) as low control (20% CP). Diets from 2 to 11 were formulated to add a mixture of synthetic amino acids including all but one of the essential amino acid to the low protein control diet to gave the total crude protein of high control diet.

The deficiency of threonine and methionine + cysteine significantly ( $P < 0.05$ ) lowered final body weight, average daily gain per fish and specific growth rate. The data showed that threonine was the first limiting amino acid, while, the second limiting amino acid was methionine + cys-

teine. The higher values of feed conversion ratio were observed for fish fed diet deficient in threonine or low control, whereas the lower value was noticed for fish fed diet high control (30% CP).

The value of protein efficiency ratio of low control diet (20% CP) was significantly ( $P < 0.05$ ) higher than the other treatments, while the higher significant values of nitrogen retention and energy retention were observed for tilapia fed high control diet and deficient of tryptophan, respectively. Body ether extract content followed the same trend to protein efficiency ratio, while the higher significant ( $P < 0.05$ ) differences of crude protein and gross energy were observed for tilapia fed high control diet. The lowest nitrogen retention in response to deduction amino acids was threonine followed in increasing order by lysine, phenylalanine + tyrosine and methionine + cysteine. Energy retention followed the same trend.

The data confirm the conclusion that, The threonine was the first limiting amino acid, whilst the second limiting amino acid was methionine + cysteine.

**Key word:** dietary essential amino acid deficiency, threonine, (methionine +cysteine), Nile tilapia ( *Oreochromis niloticus* ), performance.

## INTRODUCTION

Tilapia culture is widely practiced in many tropical and subtropical regions of the world. More than 22 tilapia species are being cultured worldwide. Nile tilapia was the 6<sup>th</sup> most cultured fin-fish species in the world in 1995 with a total production of 473,641 m.t. and an average compound growth rate of about 12% per annum since 1986 (FAO, 1997).

Amino acid requirements for fish have been determined by several methods. For many fish species, growth rates produced by diets with large amount of crystalline amino acids are inferior to diets of similar amino acid composition in which the nitrogen component is protein (Wilson *et al.*, 1978; Robinson *et al.*, 1981; Walton *et al.*, 1982, 1986 and El- Hussein *et al.*, 2001). Thus amino acid requirements obtained in this way are based on growth rates below the optimum. Ogino (1980) measured the retention of essential amino acids in the whole body protein of carp and rainbow trout and used the increase in essential amino acid con-

tent measured per periods of 14 to 28 day to estimate requirements. This method assumes that the maintenance requirement of young growing fish are low although it is not easy to reconcile this view with the fact that only 30 to 40 percent of dietary nitrogen is retained by growing fish.

Santiago and Lovell (1988) for Nile tilapia and Cowey (1994) for trout used dose-response curve method to measure the response of different amino acid levels on weight gain. Requirements of amino acid levels have been obtained from the data by broken line analysis and applying a range test by fitting a growth curve. Direct and indirect oxidation studies are also based on tissue free amino acid concentration. Kim *et al.*, (1992) used indirect oxidation studies to measure the oxidation of an essential amino acid other than the one under study, they found that as dietary concentration of the amino acid under tested increased, tissue protein synthesis will increase progressively, and the amount of other amino acids being oxidized will decrease as proportionately larger amount are used for protein synthesis.

Like other fish, tilapia require the same ten essential amino acids (EAA,s) in their diet: namely threonine, valine, methionine, isoleucine, leucine, phenylalanine, lysine, histidine, arginine and tryptophan. Santiago and Lovell (1988) analyzed weight gain of *O. niloticus* by the broken line regression method and indicated the following requirements as a percentage of the dietary protein:

Lysine, 5.12; Arginine, 4.20; Histidine, 1.72; Valine, 2.8; Leucine, 3.39; Isoleucine, 3.11; Threonine, 3.75; Tryptophan, 1.00; Methionine [with cystine 0.54% of the protein] 3.21, and Phenylalanine [with tyrosine 1.79 %of the protein] 5.54.

An optimum amino acid pattern is needed as a standard profile or reference protein when evaluating the quality of other dietary protein. (Cowy, 1994). A more restricted definition of an ideal protein is that one which includes the minimum quantity of each essential amino acid compatible with maximum utilization of the protein as a whole (Cole, 1978). No such data are available for tilapia fish.

The aim of the present study was to detect the influence of certain dietary essential amino acid deficiency on Nile tilapia fry growth performance and feed utilization.

## **MATERIALS AND METHODS**

This study was carried out in the fish Laboratory of Animal and Poultry Nutrition Department, Faculty of Agriculture, Cairo University. The study was designed to determine the effect of dietary essential amino acid deficiency on Nile tilapia fry performance and feed utilization.

### **Feed ingredients:**

Experimental diets were formulated as semi-purified diets. A mixture of casein vitamin free

and gelatin were used as the sole protein source, corn oil and cod liver oil used as the fat source; dextrin, cellulose and carboxymethyl cellulose as digestible carbohydrate, fiber and binder, respectively. All synthetic amino acid used in this study was in DL form. The optimum ratio between casein and gelatin was 5: 1 to balance amino acids pattern according to Murai *et al.*, (1984). Table (1) illustrate the proximate analysis of amino acid content of protein source (casein and gelatin ) according to A.O.A.C ( 1980).

### **Experimental fish :**

Nile tilapia fish were purchased from Nawa hatchery, Shebien EL-Kanater, Kaluobia Governorate. Fish were healthy, free from parasites with initial average weight of  $1.21 \pm 0.01$ g. The experimental fish were kept 14 days for acclimatization, thereafter , the actual experimental period followed directly from 16 June 2000 to 16 July 2000.

### **Technique of culture:**

Experiment was conducted in 12 glass aquarius of (80 x 50 x 40) cm with capacity of 160 liters representing 12 nutritional treatments. Aquaria were filled with dechlorinated tap water and supplied with aeration. Water was changed three times weekly, temperature was ranged between 26.5 -28 °C, and ammonia concentration was maintained at less than 0.3 mg /l. with pH of 8.00.



A total of 480 tilapia fry, were stocked and equally divided into 12 aquaria of 40 fish each. The recommended level of synthetic amino acid of the control diets (20 and 30 % CP) and different experimental tested diets were based on the pattern established in Santiago and Lovell (1988) ; NRC (1993) and a preceding study ( El- Hussein *et al.*, 2001). Twelve experimental diets were formulated and shown in Table (2). The first diet was formulated for high control (30% CP) and the last diet (12) was formulated as low control (20% CP). Diets from 2 to 11 were formulated to add a mixture of synthetic amino acids including all but one of the essential amino acid to the low protein control diet to give the total crude protein of high control diet (Tables 3 and 4). The twelve diets were isocaloric of 19.24 MJ/ GE /Kg and isonitrogenous of 30% crude protein according to (NRC, 1993). Fish were fed at a level of 5% of body weight twice a day at 9 a.m. and 12 a.m.

#### **Chemical composition analysis of diets and fish:**

Random samples of *O. niloticus* fry from each treatments were collected pre - and post experimental for proximate composition analysis. Dry matter, (oven dry 105°C for 24 h), Crude protein (N-Kjeldahl X 6.25), ether extract (solvent extraction with petroleum ether, bpt 40-60°C for 10-12 h), ash (oven ashed at 550°C) and crude fiber ( acid 1.25% and alkali 1.25% digestion, respectively) were analyzed according to standered methods of A.O.A.C. (1980). The gross energy

was calculated according to Hefher *et al.*, (1983). Determination of amino acids for casein and gelatin was carried out according to the methods described by Moore (1963). The oxidized samples were analyzed using a LKAB Alpha-Plus amino acid analyzer.

#### **Statistical analysis:**

The obtained data were statistically analyzed using classification one way MSTAT version 4 (1987). To investigate the relationship between carcass nitrogen retention and dietary amino acid intake data were combined to determine the linear regression. Significant differences among the mean of different treatment were carried out by Duncan's multiple range test ( $P < 0.05$ ) ( Duncan, 1955).

### **RESULTS AND DISCUSSION**

#### **1- The influence of tested dietary essential amino acids deficiency on Nile tilapia fry growth performance:**

This experiment was based on the concept that when a single amino acid is limiting in the diet, the rate of body protein accretion is directly related to that one amino acid (Cowey, 1994). However, because tilapia fry in all groups of each experiment at least doubled their body weight, the duration of experiments was sufficiently long to the quantification of traits such as deposition of protein and fat (Rodehutsord *et al.*, 1995 a and b)

Analysis of variance for data summarized in Table (5) indicated that, only the deletion of threonine and (methionine + cystine) resulted in significant ( $P < 0.05$ ) lower final body weight, relative growth rate, average daily gain per fish and specific growth rate followed in increasing order by valine, lysine, leucine, isoleucine and phenylalanine + tyrosine than the high control treatment. Therefore threonine may be considered as the first limiting amino acid, while, the second limiting amino acid was (methionine + cysteine). Santiago and Lovell (1988) showed that methionine was the first limiting amino acid

for Nile tilapia. Nordrum *et al.*, (2000) reported that sulfur containing amino acids (methionine + cysteine) seemed to be the first limiting amino acids for Atlantic salmon and methionine was less effective than cysteine in relieving the limitation. In this connection, Jackson *et al.*, (1982) reported that, threonine appeared to be limiting for tilapia and the minimum level of threonine required for growth in tilapia is lower than for carp. When diets are supplemented with all indispensable amino acids other than the first limiting amino acid or with the second most limiting amino acid this leads to a fall in the concentration of the first

Table (1): The amino acid composition of casein and gelatin (g / 100 g) used. in the experimental diets.

| Amino Acid    | Casein  | Gelatin |
|---------------|---------|---------|
| Arginine      | 3.59    | 7.12    |
| Histidine     | 2.73    | 0.95    |
| Isoleucine    | 4.30    | 1.24    |
| Leucine       | 8.54    | 2.81    |
| Lysine        | 7.31    | 3.52    |
| Methionine    | 2.69    | 0.78    |
| Cysteine      | 0.37    | 0.15    |
| Phenylalanine | 4.89    | 1.94    |
| Tyrosine      | 5.39    | 0.63    |
| Threonine     | 3.87    | 1.61    |
| Tryptophan    | *(1.19) | *(0.05) |
| Valanine      | 5.28    | 2.24    |
| Alanine       | 3.14    | 10.17   |
| Aspartic acid | 6.62    | 5.45    |
| Glutamic acid | 21.95   | 9.58    |
| Glycine       | 1.82    | 31.95   |
| Proline       | 10.85   | 16.97   |
| Serine        | 5.56    | 3.12    |

\* According to NRC (1993)

Table (2): Chemical composition of the different experimental

| Treatment assignme                   | High Control | High Control- (Arg.) | High Control- (His.) | High Control- (Iso.) | High Control- (Leu.) | High Control- (Lys.) | High Control- (Meth+ Cys.) | High Control- (Phe+ Tyr.) | High Control- (Thr.) | High Control- (Tyr.) | High Control- (Val.) | Low Control- |
|--------------------------------------|--------------|----------------------|----------------------|----------------------|----------------------|----------------------|----------------------------|---------------------------|----------------------|----------------------|----------------------|--------------|
| Ingredient %                         |              |                      |                      |                      |                      |                      |                            |                           |                      |                      |                      |              |
| Mixture A-A <sup>1</sup>             | -            | 9.62                 | 9.71                 | 9.54                 | 9.00                 | 9.22                 | 9.67                       | 8.92                      | 9.59                 | 9.87                 | 9.35                 | -            |
| C.M.C <sup>2</sup>                   | 2.00         | 2.00                 | 2.00                 | 2.00                 | 2.00                 | 2.00                 | 2.00                       | 2.00                      | 2.00                 | 2.00                 | 2.00                 | 2.00         |
| Casein                               | 26.57        | 15.94                | 15.94                | 15.94                | 15.94                | 15.94                | 15.49                      | 15.49                     | 15.49                | 15.49                | 15.49                | 15.49        |
| Gelatin                              | 5.12         | 5.12                 | 5.12                 | 5.12                 | 5.12                 | 5.12                 | 5.12                       | 5.12                      | 5.12                 | 5.12                 | 5.12                 | 5.12         |
| Dextrin                              | 38.03        | 38.41                | 38.32                | 38.49                | 39.03                | 38.81                | 38.36                      | 39.11                     | 38.44                | 38.16                | 38.68                | 48.66        |
| $\alpha$ -Cellulose                  | 17.78        | 17.78                | 17.78                | 17.78                | 17.78                | 17.78                | 17.78                      | 17.78                     | 17.78                | 17.78                | 17.78                | 17.78        |
| Corn oil                             | 2.50         | 2.50                 | 2.50                 | 2.50                 | 2.50                 | 2.50                 | 2.50                       | 2.50                      | 2.50                 | 2.50                 | 2.50                 | 2.50         |
| Cod liver oil                        | 2.50         | 2.50                 | 2.50                 | 2.50                 | 2.50                 | 2.50                 | 2.50                       | 2.50                      | 2.50                 | 2.50                 | 2.50                 | 2.50         |
| (Min-Vit) Mix.                       | 5.50         | 5.50                 | 5.50                 | 5.50                 | 5.50                 | 5.50                 | 5.50                       | 5.50                      | 5.50                 | 5.50                 | 5.50                 | 5.50         |
| Nutrient Composition (DM basis %)    |              |                      |                      |                      |                      |                      |                            |                           |                      |                      |                      |              |
| Dry matter                           | 89.69        | 89.76                | 89.61                | 89.46                | 89.73                | 89.63                | 89.39                      | 89.75                     | 89.81                | 89.66                | 89.84                | 89.65        |
| Gross Energy K.cal / Kg <sup>3</sup> | 47.11        | 4701                 | 4707                 | 4701                 | 4708                 | 4703                 | 4709                       | 4707                      | 4702                 | 4704                 | 4700                 | 4591         |
| Crude protein%                       | 30.84        | 30.71                | 30.65                | 30.44                | 30.79                | 30.61                | 30.47                      | 30.53                     | 30.67                | 30.83                | 30.39                | 20.45        |
| Ether-Extract%                       | 5.34         | 5.23                 | 5.32                 | 5.29                 | 5.36                 | 5.27                 | 5.41                       | 5.37                      | 5.26                 | 5.22                 | 5.33                 | 6.69         |
| Carbohydrate% <sup>4</sup>           | 58.66        | 58.85                | 58.88                | 59.08                | 58.62                | 58.94                | 58.96                      | 58.93                     | 58.85                | 58.78                | 59.02                | 66.74        |
| Ash%                                 | 5.16         | 5.21                 | 5.15                 | 5.19                 | 5.23                 | 5.16                 | 5.8                        | 5.17                      | 5.22                 | 5.17                 | 5.26                 | 6.12         |

<sup>1</sup> Mixture of synthetic amino acid see table (7)<sup>2</sup> Carboxymethyl Cellulose.<sup>3</sup> According to Hether *et al.* (1983).<sup>4</sup> Calculated by difference

Table (3): Mixture of synthetic amino acids supplementation the different experimental diets.

| Amino Acid    | High Control | High Control- (Arg.) | High Control- (His.) | High Control- (Iso.) | High Control- (Leu.) | High Control- (Lys.) | High Control- (Meth.+ Cys.) | High Control- (Phe.+ Tyr.) | High Control- (Thr.) | High Control- (Tyr.) | High Control- (Val.) | Low Control |
|---------------|--------------|----------------------|----------------------|----------------------|----------------------|----------------------|-----------------------------|----------------------------|----------------------|----------------------|----------------------|-------------|
| Arginine      | -            | -                    | 0.382                | 0.382                | 0.382                | 0.382                | 0.382                       | 0.382                      | 0.382                | 0.382                | 0.382                | -           |
| Histidine     | -            | 0.290                | -                    | 0.290                | 0.290                | 0.290                | 0.290                       | 0.290                      | 0.290                | 0.290                | 0.290                | -           |
| Isoleucine    | -            | 0.458                | 0.458                | -                    | 0.458                | 0.458                | 0.458                       | 0.458                      | 0.458                | 0.458                | 0.458                | -           |
| Leucine       | -            | 0.910                | 0.910                | 0.910                | -                    | 0.910                | 0.910                       | 0.910                      | 0.910                | 0.910                | 0.910                | -           |
| Lysine        | -            | 0.778                | 0.778                | 0.778                | 0.778                | -                    | 0.778                       | 0.778                      | 0.778                | 0.778                | 0.778                | -           |
| Methionine    | -            | 0.288                | 0.288                | 0.288                | 0.288                | 0.288                | -                           | 0.288                      | 0.288                | 0.288                | 0.288                | -           |
| Cysteine      | -            | 0.038                | 0.038                | 0.038                | 0.038                | 0.038                | -                           | 0.038                      | 0.038                | 0.038                | 0.038                | -           |
| Phenylalanine | -            | 0.502                | 0.502                | 0.502                | 0.502                | 0.502                | 0.502                       | -                          | 0.502                | 0.502                | 0.502                | -           |
| Tyrosine      | -            | 0.574                | 0.574                | 0.574                | 0.574                | 0.574                | 0.574                       | -                          | 0.574                | 0.574                | 0.574                | -           |
| Threonine     | -            | 0.412                | 0.412                | 0.412                | 0.412                | 0.412                | 0.412                       | 0.412                      | -                    | 0.412                | 0.412                | -           |
| Tryptophan    | -            | 0.126                | 0.126                | 0.126                | 0.126                | 0.126                | 0.126                       | 0.126                      | 0.126                | -                    | 0.126                | -           |
| Valine        | -            | 0.562                | 0.562                | 0.562                | 0.562                | 0.562                | 0.562                       | 0.562                      | 0.562                | 0.562                | -                    | -           |
| Alanine       | -            | 0.334                | 0.334                | 0.334                | 0.334                | 0.334                | 0.334                       | 0.334                      | 0.334                | 0.334                | 0.334                | -           |
| Aspartic acid | -            | 0.704                | 0.704                | 0.704                | 0.704                | 0.704                | 0.704                       | 0.704                      | 0.704                | 0.704                | 0.704                | -           |
| Glutamic acid | -            | 2.152                | 2.152                | 2.152                | 2.152                | 2.152                | 2.152                       | 2.152                      | 2.152                | 2.152                | 2.152                | -           |
| Glycine       | -            | 0.194                | 0.194                | 0.194                | 0.194                | 0.194                | 0.194                       | 0.194                      | 0.194                | 0.194                | 0.194                | -           |
| Proline       | -            | 1.154                | 1.154                | 1.154                | 1.154                | 1.154                | 1.154                       | 1.154                      | 1.154                | 1.154                | 1.154                | -           |
| Serine        | -            | 0.592                | 0.592                | 0.592                | 0.592                | 0.592                | 0.592                       | 0.592                      | 0.592                | 0.592                | 0.592                | -           |



Table (4): The total dietary amino acid content and synthetic amino acid added to the experimental diets

| Amino Acid    | High Control | High Control- (Arg.) | High Control- (His.) | High Control- (Iso.) | High Control- (Leu) | High Control- (Lys.) | High Control- (Meth.+ Cys.) | High Control- (Phe+ Tyr.) | High Control- (Thr.) | High Control- (Tyr.) | High Control- (Val.) | Low Control- |
|---------------|--------------|----------------------|----------------------|----------------------|---------------------|----------------------|-----------------------------|---------------------------|----------------------|----------------------|----------------------|--------------|
| Arginine      | 1.319        | 0.937                | 1.319                | 1.319                | 1.319               | 1.319                | 1.319                       | 1.319                     | 1.319                | 1.319                | 1.319                | 0.937        |
| Histidine     | 0.774        | 0.774                | 0.484                | 0.774                | 0.774               | 0.774                | 0.774                       | 0.774                     | 0.774                | 0.774                | 0.774                | 0.484        |
| Isoleucine    | 1.206        | 1.206                | 1.206                | 0.748                | 1.206               | 1.206                | 1.206                       | 1.206                     | 1.206                | 1.206                | 1.206                | 0.748        |
| Leucine       | 2.414        | 2.414                | 2.414                | 2.414                | 1.504               | 2.414                | 2.414                       | 2.414                     | 2.414                | 2.414                | 2.414                | 1.504        |
| Lysine        | 2.122        | 2.122                | 2.122                | 2.122                | 2.122               | 1.344                | 2.122                       | 2.122                     | 2.122                | 2.122                | 2.122                | 1.344        |
| Methionine    | 0.755        | 0.755                | 0.755                | 0.755                | 0.755               | 0.755                | 0.467                       | 0.755                     | 0.755                | 0.755                | 0.755                | 0.467        |
| Cysteine      | 0.105        | 0.105                | 0.105                | 0.105                | 0.105               | 0.105                | 0.067                       | 0.105                     | 0.105                | 0.105                | 0.105                | 0.067        |
| Phenylalanine | 1.389        | 1.389                | 1.389                | 1.389                | 1.389               | 1.389                | 1.389                       | 0.887                     | 1.389                | 1.389                | 1.389                | 0.886        |
| Tyrosine      | 1.464        | 1.464                | 1.464                | 1.464                | 1.464               | 1.464                | 1.464                       | 0.890                     | 1.464                | 1.464                | 1.464                | 0.890        |
| Threonine     | 1.110        | 1.110                | 1.110                | 1.110                | 1.110               | 1.110                | 1.110                       | 1.110                     | 0.698                | 1.110                | 1.110                | 0.698        |
| Tryptophan    | 0.318        | 0.318                | 0.318                | 0.318                | 0.318               | 0.318                | 0.318                       | 0.318                     | 0.318                | 0.142                | 0.318                | 0.192        |
| Valine        | 1.518        | 1.518                | 1.518                | 1.518                | 1.518               | 1.518                | 1.518                       | 1.518                     | 1.518                | 1.518                | 0.675                | 0.956        |
| Alanine       | 1.355        | 1.355                | 1.355                | 1.355                | 1.355               | 1.355                | 1.355                       | 1.355                     | 1.355                | 1.355                | 1.355                | 1.021        |
| Aspartic acid | 2.038        | 2.038                | 2.038                | 2.038                | 2.038               | 2.038                | 2.038                       | 2.038                     | 2.038                | 2.038                | 2.038                | 1.334        |
| Glutamic acid | 5.960        | 5.960                | 5.960                | 5.960                | 5.960               | 5.960                | 5.960                       | 5.960                     | 5.960                | 5.960                | 5.960                | 3.988        |
| Glycine       | 2.120        | 2.120                | 2.120                | 2.120                | 2.120               | 2.120                | 2.120                       | 2.120                     | 2.120                | 2.120                | 2.120                | 1.926        |
| Proline       | 3.752        | 3.752                | 3.752                | 3.752                | 3.752               | 3.752                | 3.752                       | 3.752                     | 3.752                | 3.752                | 3.752                | 2.5          |
| Serine        | 1.637        | 1.637                | 1.637                | 1.637                | 1.637               | 1.637                | 1.637                       | 1.637                     | 1.637                | 1.637                | 1.637                | 1.045        |



High L  
Control Co  
Val

1.382

2.981

4.58

Table (5): The performance of tilapia fed the different experimental diet

| Item  | High Control       | High Control- (Arg.) | High Control- (His.) | High Control- (Iso.) | High Control- (Leu.) | High Control- (Lys.) | High Control- (Meth+ Cys.) | High Control- (Phe+ Tyr.) | High Control- (Thr.) | High Control- (Try.) | High Control- (Val.) | Low Control- 1     | SE    |
|---|--------------------|----------------------|----------------------|----------------------|----------------------|----------------------|----------------------------|---------------------------|----------------------|----------------------|----------------------|--------------------|-------|
| Initial live body weight (g.)               | 1.22               | 1.21                 | 1.21                 | 1.22                 | 1.21                 | 1.21                 | 1.22                       | 1.21                      | 1.21                 | 1.21                 | 1.21                 | 1.21               | -     |
| Final live body weight (g.)                 | 2.687 <sup>a</sup> | 2.687 <sup>ab</sup>  | 2.680 <sup>ab</sup>  | 2.583 <sup>acb</sup> | 2.567 <sup>cb</sup>  | 2.537 <sup>d</sup>   | 2.463 <sup>e</sup>         | 2.610 <sup>bc</sup>       | 2.420 <sup>e</sup>   | 2.640 <sup>cb</sup>  | 2.520 <sup>d</sup>   | 2.210 <sup>f</sup> | 0.018 |
| Average daily gain (mg/day) <sup>1</sup>    | 48.89 <sup>a</sup> | 48.89 <sup>a</sup>   | 48.33 <sup>a</sup>   | 48.44 <sup>c</sup>   | 48.33 <sup>c</sup>   | 44.22 <sup>d</sup>   | 40.44 <sup>e</sup>         | 46.66 <sup>b</sup>        | 40.33 <sup>e</sup>   | 47.66 <sup>b</sup>   | 43.66 <sup>d</sup>   | 33.33 <sup>f</sup> | 0.345 |
| Specific growth rate (g./day) <sup>2</sup>  | 2.630 <sup>a</sup> | 2.630 <sup>ab</sup>  | 2.587 <sup>ab</sup>  | 2.500 <sup>cd</sup>  | 2.507 <sup>c</sup>   | 2.467 <sup>cd</sup>  | 2.297 <sup>e</sup>         | 2.560 <sup>b</sup>        | 2.307 <sup>e</sup>   | 2.600 <sup>cb</sup>  | 2.447 <sup>d</sup>   | 2.000 <sup>f</sup> | 0.056 |
| Feed consumption (g./fish)                  | 2.08               | 2.03                 | 2.08                 | 2.07                 | 1.97                 | 2.05                 | 1.95                       | 2.06                      | 1.97                 | 2.08                 | 2.05                 | 1.81               | -     |
| Feed conversion ratio per fish <sup>3</sup> | 1.413 <sup>f</sup> | 1.453 <sup>f</sup>   | 1.453 <sup>f</sup>   | 1.517 <sup>de</sup>  | 1.450 <sup>f</sup>   | 1.540 <sup>d</sup>   | 1.607 <sup>b</sup>         | 1.470 <sup>ef</sup>       | 1.627 <sup>b</sup>   | 1.4450 <sup>f</sup>  | 1.563 <sup>cd</sup>  | 1.810 <sup>a</sup> | 0.018 |

1 Average daily gain (A.D.G.) =  $1000 \times [( \text{Final body weight (g.)} - \text{Initial body weight (g.)} ) / \text{period (days)}]$

2 Specific growth rate (S.G.R.) =  $100 \times [(\ln \text{ final body weight} - \ln \text{ initial body weight}) / \text{period (days)}]$

3 Feed conversion ratio (F.C.R.) =  $\text{Dry weight of feed (g.)} / \text{weight gain (g.)}$

SE = standard error. Calculated from residual mean square in the analysis of variance.

a,b,c, etc. means in same row, with different superscripts are significantly ( $P < 0.05$ ).

Table (6): The protein in efficiency ratio protein consumption, nitrogen retention, energy intake and energy retention of tilapia fed the different experimental diet

| Item                        | High Control       | High Control- (Arg.) | High Control- (His.) | High Control- (Iso.) | High Control- (Leu.) | High Control- (Lys.) | High Control- (Meth.+ Cys.) | High Control- (Phe+ Tyr.) | High Control- (Thr.) | High Control- (Try.) | High Control- (Val.) | Low Control-       | SE±    |
|-----------------------------|--------------------|----------------------|----------------------|----------------------|----------------------|----------------------|-----------------------------|---------------------------|----------------------|----------------------|----------------------|--------------------|--------|
| Protein consumption         | 0.64               | 0.63                 | 0.63                 | 0.63                 | 0.61                 | 0.62                 | 0.59                        | 0.63                      | 0.60                 | 0.64                 | 0.62                 | 0.37               | -      |
| Protein efficiency ratio    | 2.290 <sup>b</sup> | 2.26 <sup>bc</sup>   | 2.25 <sup>bc</sup>   | 2.16 <sup>d</sup>    | 2.23 <sup>e</sup>    | 2.14 <sup>d</sup>    | 2.05 <sup>e</sup>           | 2.22 <sup>e</sup>         | 2.01 <sup>e</sup>    | 2.23 <sup>bc</sup>   | 2.11 <sup>e</sup>    | 2.70 <sup>a</sup>  | 0.053  |
| N-Retention                 | 43.77 <sup>b</sup> | 40.34 <sup>c</sup>   | 36.61 <sup>e</sup>   | 34.37 <sup>f</sup>   | 37.24 <sup>e</sup>   | 30.43 <sup>h</sup>   | 34.35 <sup>f</sup>          | 33.58 <sup>g</sup>        | 27.94 <sup>i</sup>   | 44.61 <sup>e</sup>   | 40.13 <sup>c</sup>   | 38.05 <sup>d</sup> | 0.707  |
| Energy Intake (Kcal / fish) | 9.79               | 9.73                 | 9.69                 | 9.73                 | 9.27                 | 9.64                 | 9.18                        | 9.69                      | 9.26                 | 9.78                 | 9.63                 | 8.30               | -      |
| Energy Retention            | 26.26 <sup>a</sup> | 22.75 <sup>d</sup>   | 21.98 <sup>f</sup>   | 21.63 <sup>g</sup>   | 23.77 <sup>e</sup>   | 19.93 <sup>h</sup>   | 21.81 <sup>g</sup>          | 22.22 <sup>ef</sup>       | 18.93 <sup>i</sup>   | 25.21 <sup>b</sup>   | 22.48 <sup>de</sup>  | 19.28 <sup>i</sup> | 0.1445 |

SE = standard error. Calculated from residual mean square in the analysis of variance.

a,b,c... etc. means in same row with different superscripts are significantly (P 0.05).

Table (7): The whole body composition of tilapia fed the different experimental diet

| Item                  | High Control        | High Control- (Arg.) | High Control- (His.) | High Control- (Iso.) | High Control- (Leu.) | High Control- (Lys.) | High Control- (Meth+ Cys.) | High Control- (Phe+ Tyr.) | High Control- (Thr.) | High Control- (Try.) | High Control- (Val.) | Low Control-         | SE±    |
|-----------------------|---------------------|----------------------|----------------------|----------------------|----------------------|----------------------|----------------------------|---------------------------|----------------------|----------------------|----------------------|----------------------|--------|
| Moisture              | 73.14 <sup>bc</sup> | 73.20 <sup>bed</sup> | 73.17 <sup>ab</sup>  | 73.21 <sup>ab</sup>  | 74.41 <sup>abc</sup> | 75.56 <sup>a</sup>   | 73.89 <sup>cde</sup>       | 74.11 <sup>bcd</sup>      | 75.32 <sup>ab</sup>  | 72.79 <sup>c</sup>   | 73.54 <sup>cde</sup> | 74.13 <sup>bcd</sup> | 1.071  |
| Crude protein %       | 63.25 <sup>a</sup>  | 62.35 <sup>a</sup>   | 61.05 <sup>b</sup>   | 60.11 <sup>b</sup>   | 60.17 <sup>b</sup>   | 57.53 <sup>c</sup>   | 58.50 <sup>c</sup>         | 56.17 <sup>d</sup>        | 56.20 <sup>d</sup>   | 63.11 <sup>a</sup>   | 62.51 <sup>a</sup>   | 53.98                | 1.242  |
| Ether extract %       | 22.27 <sup>b</sup>  | 20.71 <sup>c</sup>   | 22.41 <sup>d</sup>   | 23.84 <sup>b</sup>   | 23.91 <sup>b</sup>   | 24.21 <sup>b</sup>   | 22.97 <sup>c</sup>         | 23.88 <sup>b</sup>        | 24.20 <sup>b</sup>   | 20.91 <sup>c</sup>   | 20.92 <sup>c</sup>   | 25.11 <sup>a</sup>   | 0.4471 |
| Ash %                 | 15.10 <sup>f</sup>  | 16.70 <sup>d</sup>   | 16.08 <sup>e</sup>   | 15.83 <sup>e</sup>   | 15.35 <sup>ef</sup>  | 17.97 <sup>e</sup>   | 18.23 <sup>e</sup>         | 19.71 <sup>a</sup>        | 19.10 <sup>b</sup>   | 15.75 <sup>e</sup>   | 16.02 <sup>e</sup>   | 19.98 <sup>a</sup>   | 0.554  |
| Energy Kcal/100g fish | 567 <sup>a</sup>    | 549 <sup>de</sup>    | 559 <sup>b</sup>     | 566 <sup>a</sup>     | 567 <sup>a</sup>     | 555 <sup>bc</sup>    | 458 <sup>f</sup>           | 544 <sup>e</sup>          | 548 <sup>de</sup>    | 555 <sup>bc</sup>    | 553 <sup>cd</sup>    | 546 <sup>e</sup>     | 4.840  |

SE = standard error. Calculated from residual mean square in the analysis of variance.

a, b, c, etc. means in same row with different superscripts are significantly (P &lt; 0.05).



limiting amino acid in the blood and eventually to reduce feed intake (NRC, 1993). The same trend was observed for reduced feed consumption of tilapia as reaction to omitting (20%) of dietary threonine and methionine + cysteine. The data agreed with the findings of Cho *et al.*, (1992) for arginine, Cowey *et al.*, (1992) for methionine and Pfeffer *et al.*, (1992) for lysine. The experiment indicated that the values of feed conversion ratio (Table, 5) was ranged between 1.41 and 1.81. The higher values of feed conversion ratio ( low feed efficiency value ) were observed for fish fed diet omitted in threonine and low control diet, the lower values was observed for fish fed diet high control diet (high feed efficiency value).

#### **Feed utilization and tilapia body composition:**

Protein efficiency ratio; nitrogen retention and energy retention are presented in (Table, 6) .The higher significant ( $P < 0.05$ ) values were observed for fish fed the low control diet (20% CP) than the other treatments (30% CP). Ether extract content followed the same trend, while, the opposite trend was noticed in body crude protein content and body gross energy content (Table, 7). The present data indicated that among the traits monitored, nitrogen retention (protein deposition) is the most sensitive indicator of sub-optimal supply of an amino acid. This conclusion could be drawn from previous results previously for methionine, arginine and threonine (Rodehutsord *et al.*, 1995 a and b)

The change in nitrogen retention among treatments when certain amino acids were omitted from the amino acids mixture in the dietary casein protein pattern, suggesting that imbalances between essential amino acids were sufficient to depress nitrogen retention (Table,6). Tryptophan was the only amino acid when removed from the protein pattern nitrogen retention improved. This result may be unacceptable due to the level of tryptophan requirement in this experiment obtained from NRC (1993) and not determine. The deduction of arginine and valine had a little effect, whereas, the lowest nitrogen retention in response to deduced amino acids was threonine followed by lysine, phenylalanine + tyrosine and methionine + cystine in increasing order. Energy retention followed the same trend. The equation of the linear regression relating body weight and body nitrogen retention to intake of the essential amino acid was shown in Table (8). Hephner (1988) reported that a few amino acid such as leucine and isoleucine from carbon skeleton, which yield intermediate metabolites experimental diets that are more closely related to the metabolism of fatty acids than to that of carbohydrate such as acetate, acetoacetate,  $\beta$ -hydroxybutyrate and acetone. Eventually these produce acetyl-CoA which may be either utilized for synthesis of fatty acids or oxidized for production of energy through the TCA cycle. The data reported herein (Table, 7) indicated that for body ether extract of tilapia fry fed diet omitted for leucine (23.91%) and isoleucine (23.84%) were higher than fish fed

Table 8: The equation of the linear regression relating body weight and body nitrogen retention of each dietary essential amino acid.

| Amino acid               | Body weight equation             | r     | SE*     |
|--------------------------|----------------------------------|-------|---------|
| Arginine                 | $Y = 2.575 + 3.750 X$            | 0.509 | 3.172   |
| Histidine                | $Y = 2.545 + 8.750 X$            | 0.945 | 1.510   |
| Isoleucine               | $Y = 2.447 + 9.375 X$            | 0.911 | 2.124   |
| Leucine                  | $Y = 2.400 + 5.661 X$            | 0.948 | 0.955   |
| Lysine                   | $Y = 2.318 + 8.280 X$            | 0.928 | 1.659   |
| Methionine + Cysteine    | $Y = 2.202 + 25.603 X$           | 0.884 | 6.753   |
| Phenylalanine + Tyrosine | $Y = 2.495 + 3.153 X$            | 0.904 | 0.745   |
| Threonine                | $Y = 2.072 + 26.036 X$           | 0.932 | 5.067   |
| Tryptophan               | $Y = 2.606 + 10.508 X$           | 0.910 | 2.393   |
| Valine                   | $Y = 2.390 + 9.265 X$            | 0.997 | 0.370   |
|                          | Body nitrogen retention equation |       |         |
| Arginine                 | $Y = 34.19 + 341.79 X$           | 0.848 | 106.67  |
| Histidine                | $Y = 26.82 + 1028.59 X$          | 0.936 | 193.55  |
| Isoleucine               | $Y = 21.64 + 871.69 X$           | 0.956 | 133.468 |
| Leucine                  | $Y = 27.98 + 338.63 X$           | 0.868 | 96.891  |
| Lysine                   | $Y = 10.63 + 745.62 X$           | 0.969 | 94.28   |
| Methionine + Cysteine    | $Y = 25.65 + 957.5 X$            | 0.892 | 243.14  |
| Phenylalanine + Tyrosine | $Y = 18.19 + 422.34 X$           | 0.973 | 50.549  |
| Threonine                | $Y = 7.20 + 1548.51 X$           | 0.936 | 290.71  |
| Tryptophan               | $Y = 44.79 + 36.95 X$            | 0.160 | 114.09  |
| Valine                   | $Y = 37.37 + 198.39 X$           | 0.989 | 14.64   |

\* Standard error of slop

gh control (22.27%). The data agreed with those obtained by Hepher (1988).

enerally, some adverse interactions may occur between amino acids that are structurally related when their concentrations in the diet are imbalance. Recently, well-known examples of fish are antagonisms arising from dietary imbalance of lysine-arginine and of leucine-valine (Lall *et al.*, 1994; Nordum *et al.*, 2000 ). No data are available

on such interaction in tilapia. In the present study, no consistent relationship between dietary amino acids was detected.

To use the results as a reference pattern for dietary amino acids in evaluating the quality of dietary proteins and for practical diet, the formulation and amino acid digestibility must also be considered and maintenance requirement.

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