

A COMPARATIVE STUDY BETWEEN METHODS USED TO EVALUATE GROSS ENERGY IN FEED, FISH AND FAECAL MATTER

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SAMMARY

Three different methods were used to determine and compare between and gross energy content of feed, fish-edible parts and faecal samples. method 1: the GE content was determined from the data of the actual chemical composition and the caloric values of the energy bearing nutrients; viz 5.65 , 9.45 and 4.15 Kcal /g for protein, fat and carbohydrate, respectively. Method 2: the GE content was determined by combustion of the concerned materials in bomb calorimeter. Method 3: the GE content was determined by dichromate wet oxidation. The results indicated that combustion is the most accurate method for determination of GE content of the three tested materials. Calculation using chemical composition and determination by wet dichromate oxidation underestimated the GE content. Justification of the results could be reached by using a factor 1.02-1.05; with an average of 1.035 for appropriation and simplification. Further investigations are, however, required for a greater level of accuracy of the assigned factor.

INTRODUCTION

Energy is required for all phases of body metabolism. In the overall subject of animal nutrition, bioenergetics is very important, because energy is quantitatively the most important item in animal diets and all animal feeding standards are based mainly on energy needs. With all other nutrients, modern laboratory procedures allow us to fractionate feedstuff, animal tissue and so forth into their compositional component, so protein, lipid, vitamins and minerals can be segregated. However, study of energy metabolism requires a different approach because energy may be derived from most organic compounds ingested by the animal.

Gross energy (GE) values oftenly obtained on feedstuffs or diets in the process of arriving to energy utilization, GE is the quantity of heat resulting from the complete oxidation of feed or other substances (Church and Pond, 1988). The nutritive value of supplementary feeds for fish depends on the energy content and the digestibility of feed components.

Methods developed to measure the different components of energy are used in feed evaluation research and energy requirements. GE of feed, fish and faecal matter can be determined by different methods (Craig et al., 1978). It can be directly obtained by bomb calorimetry, or calculated from chemical composition and it can be calculated from data obtained through the chemical oxygen demand (COD) method. The choice of one of the three methods depends upon the equipment available, time needed, costs involved and/or the amount of sample obtained. The currently available results are greatly conflicting; Therefore the present study was planned to evaluate and compare between these three different methods used as means for gross energy determination in feed, fish and faecal matter. The obtained data may be used as a guide in evaluation of newly introduced feedstuffs in fish diets.

MATERIAL AND METHODS

This work was carried out in the Fish Culture Research Station at El-Kanater El-Khyria, where 60 Tilapia fingerlings with an average weight of 25.6 ± 2.7 g were kept in four glass aquaria (15 each) supplied with dechlorinated tap water and fed on the experimental diet formulated to cover their requirements (table 1). The fish were fed every day at a rate of 5% of live body weight, the faeces were removed every day using a siphoning

tube and fine mash (20 u) and transferred to petri dishes for drying, the experiment lasted for 20 days.

Twenty samples were equally obtained from feed and fish (edible parts) and ten samples of faecal matter, all samples were analyzed for dry matter (DM), crude protein (CP), fat (EE) and ash content. The samples were homogenized in a mincing machine and / or coffee- grinder, and analyzed according to A. O. A. C. (1980).

Determination of gross energy content:

The gross energy content of the dry matter of feed, fish and faecal samples was determined by the three following methods:

* First method: The GE content was calculated using values of 5.65, 9.45 and 4.15 Kcal/g of the chemically analysed protein, fat and carbohydrate, respectively.

* Second method: The GE content was determined by combustion of one gram of the sample in a bomb calorimeter according to Nijkamp (1969 and 1971).

* Third method: The GE content was determined by wet oxidation for two hours at 148 C° of about 1.5 mg of sample in 2ml distilled water plus 2 ml of reaction mixture. The reaction mixture was prepared by dissolving 6.129 gm K₂ Cr₂ O₇ in 60ml distilled water and making up to 500 ml with concentrated H₂SO₄ containing 10 g Ag SO₄ per liter. The chemical oxygen demand (COD) was derived from the spectrophotometrically measured difference at 623 nm between tested samples and blanks. The (COD) was converted to energy using a factor of 3.4 (Macioleu 1962), the energy data were subsequently corrected for incomplete oxidation of protein, whereby correction was done by adding 5.94 Kcal/g NH₃-N or 0.95 Kcal / g protein (Elliott and Davison 1975).

Statistical analysis

It was adopted according to Steel and Torrie (1980) using the factorial analysis of variance.

Duncan's test was applied in each experiment whenever possible to test mean differences (Duncan, 1955).

RESULTS AND DISCUSSION

Twenty feed and twenty fish samples as well as ten faecal matter samples were subjected to three methods for estimation of the GE. A comparative evaluation of the results was statistically analyzed.

Results of chemical analysis of fish edible parts are shown in table (2) while that of faecal matter presented in table (3). Data obtained from the present study indicated that differences in the methods used to determine dry matter gross energy content of diet, fish and faecal matter

Table (1) : composition of the experimental diet.

Ingredient	%
Soybean meal (44% cp)	30
Yellow corn (9% cp)	24
Wheat bran (12% cp)	20
Fish meal (60 % cp)	15
Meat meal (54.1 % cp)	6
Cotton seed oil	3
Vitamin *	1.0
mineral premix **	1.0
Chemical composition %	
Moisture	10.10
Crude protein	29.81
Ether extract	6.87
Crude fiber	5.95
Ash	8.26
Nitrogen free extract	39.01

* Each kg of mineral premix contains Ca 125 gm, P 90 gm, Fe 25000 mg, Cu 2000 mg, Mn 60000 mg, I 200 mg. Se 100mg, Zn 40000 mg and NaCl 250 mg.

** Each kg of vitamin premix contains Vit. A 4000.000 I. U., Vit D 8000 I. U., Vit. e 1000 mg, Vit B1 2000 mg; B2 10000 mg, B3 10000 mg, B6 100 mg Vit. K 1000 mg, B12 3mg, Vit. C 10 mg, folic acid 500 mg, Pantothenic acid 5000 mg and Niacine amide 5000 mg.

(Table 4) were significant different ($P < 0.05$). The data of gross energy content of feed obtained by combustion in a bomb calorimeter, chemical composition (calculation) and by chemical oxygen demand (COD) were 4480.43, 4396.89 and 4281.39 Kcal / g, respectively.

Table (2): Analyzed (protein and fat) and calculated (Carbohydrate) and dry matter composition (%) of the fish edible parts ($n = 20 \pm$ SD).

Item	%
Crude protein	62.95 \pm 2.90
Ether extract	22.03 \pm 1.85
Crude fiber *	-- \pm --
Ash	13.42 \pm 1.21
Nitrogen free extract	1.60 \pm 0.11

* Assuming zero percent.

Table (3): Average chemical composition of the faecal matter of fish fed the experimental diet ($n = 10 \pm$ SD).

Item	%
Crude protein	19.80 \pm 1.23
Ether extract	6.23 \pm 0.85
Crude fiber	11.53 \pm 2.35
Ash	13.56 \pm 2.61
Nitrogen free extract	38.88 \pm 4.78

Table (5): Results of the analysis of variance on dry matter gross energy content of the feed, fish and faeces sample, the model contained the material investigated, i.e. feed, fish or faeces, as a main factor wherein the effect of method applied was nested.

Source of variation	d.f.	Mean squares	F ratio	P value
Methods within feed	2	13010.86	9.70	< 0.05
Methods within fish	2	28506.85	10.11	< 0.05
Methods within faeces	2	16965.82	15.39	< 0.05

Table (4): Dry matter gross energy (Kcal/gm) of the feed, fish and faeces samples as determined with three different methods, i.e. calculated on basis of chemical composition, combusted in a bomb calorimeter and oxidized chemically.

Material	Calculated	Combusted	COD	\pm SE
Feed *	4396.89 b	4480.43 a	4281.39 c	25.89
Fish **	5705.70	5767.06	5600.16	37.54
Faeces **	3752.75	3811.05	3681.03	23.48

* Values are the average of twenty samples.

** Values are the average of ten samples.

Values of fish were 5767.06, 5705.70 and 5600.16 Kcal/g, respectively and that of the faecal matter were 3811.05, 3752.75 and 3681.03 Kcal / g, respectively.

The effect of methods applied on gross energy content was significant different ($P \leq 0.05$) for each material tested (Table 4). The data of gross energy content based on both chemical composition (calculation) and COD were significantly ($P < 0.05$) lower than those obtained by combustion in a bomb calorimeter. Values of feed were (4396.89, 4281.39 vs 4480.43 Kcal / g), of fish were (5705.70, 5600.16 vs 5767.06 Kcal/g) and that of faecal matter were (3750.75, 3681.03 vs. 3811.05 Kcal / g), respectively.

The COD method provided the lowest data for each of the three materials the results were on the average 96.42% of those obtained by combustion in a bomb calorimeter this may be due to incomplete oxidation of lipids and / or proteins; since carbohydrates are expected to be completely oxidizable by dichromate (Maciolek, 1962).

Determination of gross energy content by dichromate wet oxidation is the fastest of the three methods investigated. It also may be the only alternative remaining in the case of a small sample. However, the results obtained will need a correction to estimate the true gross energy content (by calorimeter). In the present study due to incomplete oxidation of the organic constituents multiplying by a value between 1.02 and 1.05 (with an average of 1.035) seems to be the most appropriate.

In conclusion it can be stated that combustion is the most accurate method for determination of the gross energy content of feed, fish and faecal matter. calculations from both chemical composition and determination by dichromate wet oxidation underestimate the gross energy content of the three materials tested. However, further investigations are needed to justify the above cited factor assigned for a greater level of accuracy.

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