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Digestibility of Local Feed Ingredients in Tilapia *Oreochromis niloticus* Juveniles, Determined on Faeces Collected by Siphoning or Stripping

Francis Pius Mmanda [1,2,3], Jan Erik Lindberg [1], Anna Norman Haldén [4], Matern S. P. Mtolera [2], Rukia Kitula [2] and Torbjörn Lundh [1,*]

1 Department of Animal Nutrition and Management, Swedish University of Agricultural Sciences, P.O. Box 7024, SE-75007 Uppsala, Sweden; francis.pius.mmanda@slu.se (F.P.M.); Jan.Erik.Lindberg@slu.se (J.E.L.)
2 Ministry of Livestock and Fisheries, Dodoma P.O. Box 2847, Tanzania; mtolera@yahoo.co.uk (M.S.P.M.); rkitula@yahoo.com (R.K.)
3 Institute of Marine Sciences, University of Dar es Salaam, Zanzibar P.O. Box 668, Tanzania
4 Department of Biomedical Sciences and Veterinary Public Health, Swedish University of Agricultural Sciences, P.O. Box 7028, SE-75007 Uppsala, Sweden; anna.norman-halden@slu.se
* Correspondence: Torbjorn.Lundh@slu.se

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Abstract: Eight locally available protein source ingredients in Tanzania were selected for assessment of apparent digestibility (AD) in tilapia *Oreochromis niloticus*, using faeces samples collected by siphoning or stripping. The selected protein source ingredients were Lake Victoria sardines (FM), brewers spent yeast (BSY), moringa leaves (ML), freshwater shrimp (FSH), marine shrimp (MSH), cattle blood (CB), duckweed (DW) and fish frames (FF). The AD (%) of dry matter (DM), organic matter (OM) and crude protein (CP) was unaffected ($p > 0.782$–0.901) by the faeces collection method (i.e., siphoning or stripping), with correlation coefficient (r) of 0.98, 0.99 and 0.93 between AD values for DM, OM and CP, respectively, following siphoning and stripping. The AD (%) of DM, OM, CP and gross energy (GE) in the test ingredients differed ($p < 0.0001$). The AD (%) of DM and OM was lowest in BSY and DW, followed in increasing order by ML, MSH, FF, FSH and CB. In general, the AD (%) of CP was high (>76%), but with a low value (46%) for DW. The AD (%) of GE was closely correlated (r = 0.96) with the AD of OM. In conclusion, FSH, MSH, CB, FF, BSY and ML have acceptable protein digestibility to be used in tilapia diet formulation.

Keywords: nutritive value; apparent digestibility; non-conventional ingredients; protein source; protein; energy; fish diets; Nile tilapia

1. Introduction

Aquaculture production is continuously increasing worldwide to meet global market demand for fish and fishery products, driven by the diminishing of wild capture fisheries as the result of over-population and over-exploitation [1]. The world aquaculture production in 2018 was 82 million tons and accounted for 46% of the total fish production [2]. In Tanzania, the aquaculture production in 2019/2020 was 18,717 tons of which over 90% was tilapia fish production [3]. In recent years, the number of tilapia fish farmers has gradually increased in Tanzania, resulting in a corresponding increase in demand for fish feed. Currently, the market price of commercial fish feeds available in the country is very expensive and therefore unaffordable to small-scale fish farmers. Instead, the majority of tilapia fish farmers in Tanzania rely on locally available feed ingredients for their farmed fish [4]. In general,

there is limited information available on the chemical composition and potential nutritive value of local feed ingredients.

The potential nutritive value of feed ingredients can be assessed based on the digestibility of energy and nutrient components [5,6]. The digestibility of energy and nutrients in fish has been evaluated on faeces samples collected by different methods including siphoning [7,8] and stripping [9,10], combined with the use of indigestible markers (e.g., chromic oxide and titanium dioxide) in the diet. However, there is limited published data comparing the efficacy of these fecal matter collection methods used for a digestibility assessment.

Nile tilapia (*Oreochromis niloticus*) is the most commonly cultured fish species in Tanzania [4,11,12]. The popularity of Nile tilapia is due to its market acceptability, fast growth rate, resistance to disease and ability to grow on a wide range of diets. It is also very tolerant to a wide range of environmental conditions, has the ability to reproduce readily in captivity and has a high prolific rate and good carcass taste. To reduce their production costs, in the past most tilapia fish farmers relied on locally available feed ingredients to supplement the diet of their cultured fish [4].

The evaluation of potential local feed ingredients used by tilapia fish farmers in Tanzania has been underway for several decades. Most information on the nutritive value of local feed ingredients in the country has been obtained by proximate chemical composition analysis [13–16]. Available digestibility data mainly relate to poultry [13], rabbits [17] and African catfish [15] while many studies on potential feed ingredients for fish have been reported worldwide [5,9,14,15,18–21]. Literature data on the use of alternative dietary protein sources for farmed tilapia including by-products of both animal and plant origin were evaluated in a review [22]. However, there is limited information on the nutrient content and energy digestibility of local feed ingredients for cultured tilapia in Tanzania.

The aim of the present study was thus to evaluate the chemical composition and digestibility of commonly used local feed ingredients from different geographical regions of Tanzania. In addition, the aim was to evaluate the efficacy of siphoning and stripping in the digestibility determination in Nile tilapia juveniles.

2. Results

2.1. Chemical Composition of Ingredients and Diets

The proximate chemical composition of the test ingredients was 40.4–460 g kg^{-1} dry matter (DM) for ash, 199–840 g kg^{-1} DM for crude protein (CP), 0.4–118 g kg^{-1} DM for crude fiber (CF), 28.3–151.0 g kg^{-1} DM for ether extract (EE) and 29.7–493.0 g kg^{-1} DM for nitrogen-free extract (Nfe) (Table 1). The proximate chemical composition of the other dietary ingredients (i.e., cotton seed cake, sunflower seed cake, maize bran and wheat pollard) are shown in Table 1.

Table 1. Chemical composition (g/kg DM) of the reference diet and test diet ingredients.

Test Ingredient	Ash	CP	CF	EE	Nfe
Fishmeal	223	650	2.6	80.7	43.4
Brewers spent yeast	162	390	17.1	28.3	403
Moringa leaf	118	298	52.2	38	493
Freshwater shrimp	447	468	12.5	42.5	29.7
Marine shrimp	224	605	20.9	70.9	79.2
Cattle blood	40.4	840	0.4	46.5	73.3
Duckweed	185	199	118	40.9	458
Fish frames	460	352	7.3	151	30.3
Cotton cake	87.3	204	212	84	413
Sunflower cake	215	235	251	152	148
Maize bran	61.1	124	58.1	175	582
Wheat pollard	42.4	177	47	69.2	665

CP: Crude protein; CF: crude fiber; EE: ether extract; Nfe: nitrogen-free extract.

The fixed proportion of test ingredient inclusion (30% of DM) in the experimental diets in combination with the large variation in the proximate chemical composition between test ingredients resulted in a wide range of proximate chemical compositions in the test diets (Table 2).

Table 2. Ingredients (g/kg) and chemical composition (g/kg DM) of reference diet (FMD) and test diets.

	Diet [1]							
Ingredients	**FMD**	**BSYD**	**MLD**	**FSHD**	**MSHD**	**CBD**	**DWD**	**FFD**
Fishmeal	338	237	237	237	237	237	237	237
Brewers spent yeast	-	299	-	-	-	-	-	-
Moringa leaves	-	-	299	-	-	-	-	-
Inland shrimp	-	-	-	299	-	-	-	-
Marine shrimp	-	-	-	-	299	-	-	-
Blood	-	-	-	-	-	299	-	-
Duckweed	-	-	-	-	-	-	299	-
Fish frames	-	-	-	-	-	-	-	299
Cotton cake	159	111	111	111	111	111	111	111
Sunflower cake	49.8	34.8	34.8	34.8	34.8	34.8	34.8	34.8
Maize bran	199	139	139	139	139	139	139	139
Wheat pollard	189	132	132	132	132	132	132	132
Cassava flour	29.9	20.9	20.9	20.9	20.9	20.9	20.9	20.9
Chromic oxide	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Sunflower oil	19.8	13.9	13.9	13.9	13.9	13.9	13.9	13.9
Vitamin/Mineral	10.0	7.0	7.0	7.0	7.0	7.0	7.0	7.0
Chemical Composition								
Ash	121	133	124	219	152	96.9	141	222
Crude protein	322	342	315	366	407	477	285	331
Crude fiber	68.2	52.7	63.2	51.4	53.9	47.8	82.9	49.8
Ether extract	102	76.0	78.9	80.2	88.7	81.4	79.7	113
Nitrogen-free extract	387	396	420	284	299	297	412	284

[1] FMD: fishmeal diet (reference); BSYD: brewers spent yeast diet; MLD: moringa leaf diet; FSHD: freshwater shrimp diet; MSHD: marine shrimp diet; CBD: cattle blood diet; DWD: duckweed diet; FFD: fish frames diet.

2.2. Faeces Collection Method

Siphoning: the apparent digestibility (AD) (%) of DM was highest ($p < 0.0001$) in the cattle blood diet and the freshwater shrimp diet followed by the marine shrimp and moringa leaf diets (Table 3). The AD (%) of organic matter (OM) was highest ($p < 0.0001$) in the cattle blood diet followed by the freshwater shrimp and fish frames diets. The AD (%) of CP was higher ($p < 0.0001$) in the freshwater shrimp diet than in the other diets. The AD (%) of gross energy (GE) was highest ($p < 0.0001$) in the cattle blood diet followed by the freshwater shrimp and fish frames diets.

Table 3. Apparent digestibility (AD, %) of dry matter (DM), organic matter (OM), crude protein (CP) and gross energy (GE) in the reference diet (FMD) and test diets in Nile tilapia juveniles, determined on faeces collected by siphoning or stripping.

Diet	AD_DM	AD_OM	AD_CP	AD_GE
Siphoning				
Fishmeal	25.5 ^{a}	37.7 ^{a}	76.9 ^{a}	44.4 ^{a}
Brewers spent yeast	23.5 ^{a}	29.5 ^{b}	75.8 ^{a}	37.4 ^{b}
Moringa leaf	29.8 ^{ab}	38.4 ^{a}	78.4 ^{ab}	40.8 ^{ab}
Freshwater shrimp	41.3 ^{c}	51.6 ^{c}	84.5 ^{b}	58.1 ^{c}
Marine shrimp	33.3 ^{b}	41.3 ^{a}	79.5 ^{ab}	49.2 ^{d}
Cattle blood	46.4 ^{d}	61.6 ^{d}	80.9 ^{bc}	78.0 ^{e}
Duckweed	21.6 ^{a}	27.9 ^{b}	73.1 ^{ad}	35.3 ^{b}
Fish frames	36.0 ^{b}	49.1 ^{c}	76.9 ^{a}	52.7 ^{d}
SEM	1.07	1.09	0.69	0.93
p-value	<0.0001	<0.0001	<0.0001	<0.0001
Stripping				
Fishmeal	27.6 ^{a}	39.6 ^{a}	75.5 ^{a}	-
Brewers spent yeast	24.0 ^{b}	28.8 ^{b}	76.8 ^{a}	-
Moringa leaf	28.3 ^{a}	36.2 ^{ab}	78.5 ^{ab}	-
Freshwater shrimp	40.3 ^{d}	51.3 ^{c}	83.6 ^{b}	-
Marine shrimp	32.5 ^{c}	40.3 ^{a}	82.1 ^{b}	-
Cattle blood	47.2 ^{e}	64.4 ^{d}	79.7 ^{ab}	-
Duckweed	23.6 ^{b}	30.1 ^{b}	72.4 ^{ac}	-
Fish frames	37.8 ^{d}	50.3 ^{c}	76.4 ^{ab}	-
SEM	0.81	0.87	0.76	-
p-value	<0.0001	<0.0001	<0.0001	-
Comparison of Faeces Collection Methods				
Siphoning	32.2	42.1	78.2	-
Stripping	32.6	42.6	78.1	-
SEM	1.72	2.31	0.77	-
p-value	0.782	0.828	0.901	-

Values within columns with different superscript letters are significantly different as determined by Tukey–Kramer at $p < 0.05$; SEM: standard error of the mean.

Stripping: the AD (%) of DM and OM was highest ($p < 0.0001$) in the cattle blood diet followed by the freshwater shrimp and fish frames diets (Table 3). The AD (%) of CP was higher ($p < 0.0001$) in the freshwater shrimp and marine shrimp diets than in the other diets.

Overall, the AD (%) of DM, OM and CP in the reference diet (fishmeal diet), and the test diets was unaffected ($p > 0.782$–0.901) by the faeces collection method (siphoning or stripping; Table 3). The correlation coefficient (r) between the AD of DM, OM and CP determined by stripping and siphoning was 0.98, 0.99 and 0.93, respectively (Table 4).

Table 4. Relationship between apparent digestibility (AD, %) of dry matter, organic matter and crude protein determined by stripping (y) and siphoning (x).

Item	Equation	R^2
Dry matter	y = 1.011 x	0.970
Organic matter	y = 0.985 x	0.978
Crude protein	y = 0.999 x	0.868

2.3. Digestibility of Test Ingredients

The AD (%) of DM, OM, CP and GE in the test ingredients differed ($p < 0.0001$). The AD (%) of DM and OM was lowest in duckweed and brewers spent yeast followed in increasing order by moringa leaf, marine shrimp, fish frames, freshwater shrimp and cattle blood (Table 5). The AD (%) of CP was lower in duckweed than in the other test ingredients. The AD (%) of GE was lower in duckweed and

brewers spent yeast followed by moringa leaf, marine shrimp, fish frames, freshwater shrimp and cattle blood.

Table 5. Apparent digestibility of test ingredient (AD, %) dry matter (DM), crude protein (CP), organic matter (OM) and gross energy (GE) in Nile tilapia juveniles, determined on faeces collected by siphoning or stripping.

Sample	AD_DM	AD_CP	AD_OM	AD_GE
Brewers spent yeast	16.5 ^{a}	76.9 ^{a}	7.66 ^{a}	19.6 ^{a}
Moringa leaf	35.0 ^{b}	90.3 ^{b}	34.7 ^{b}	31.5 ^{a}
Freshwater shrimp	73.8 ^{c}	110 ^{c}	93.5 ^{c}	104 ^{b}
Marine shrimp	48.9 ^{d}	92.4 ^{b}	45.8 ^{d}	61.5 ^{c}
Cattle blood	94.7 ^{e}	87.8 ^{ab}	109 ^{e}	108 ^{b}
Duckweed	13.0 ^{a}	46.3 ^{d}	7.63 ^{a}	14.0 ^{a}
Fish frames	61.0 ^{f}	78.7 ^{ab}	86.9 ^{c}	76.9 ^{d}
SEM	2.12	2.78	2.21	3.30
p-Value	<0.0001	<0.0001	<0.0001	<0.0001

Values within columns with dDouble references have been changed ifferent superscript letters are significantly different as determined by Tukey–Kramer at $p < 0.05$; SEM: standard error of the mean.

3. Discussion

The present study showed that the estimated AD of dry matter, organic matter and protein in tilapia was unaffected by the faeces collection method (i.e., siphoning or stripping) with high correlation coefficients between the methods (r = 0.93–0.99). This suggests that either of the methods may be applied in digestibility studies with tilapia and that the experimental facilities could decide the preferred method of choice. The AD of feed ingredients and fish feeds has been determined previously following faeces collection by the siphoning method [7,8] and the stripping method [9,10] but only a few studies have compared the two methods in tilapia. No significant differences in estimates of digestibility of protein, Nfe and ash using siphoning or stripping in tilapia fed soybean diets were found [7]. Weatherup and McCracken compared estimates of digestibility in rainbow trout (*Oncorhynchus mykiss*) using two markers (titanium dioxide and chromic oxide) and two methods of faeces collection (stripping and mechanical sieving of faeces from the outlet water of the fish tank; the Choubert method). They found that both methods of faeces collection had inherent weaknesses but the Choubert method provided more accurate and repeatable measurements [23]. Storebakken et al. [24] compared estimates of faeces digestibility in Atlantic salmon (*Salmo salar*) following stripping, sieving (Choubert method) and dissection and observed differences between the methods in the AD of dry matter in the ascending order of stripping < sieving < dissection.

Variations in the quality and quantity of dietary nutrients influence the digestibility and growth performance in fish [8,25]. However, the digestibility of nutrients and energy differs from one fish species to another and even within an individual fish depending on age, sex, species, water temperature and diet composition [6]. The results of the present study indicate that freshwater shrimp, marine shrimp, cattle blood, fish frames, brewers spent yeast and moringa leaves have acceptable protein digestibility to be used in tilapia diet formulation. The low energy digestibility observed for duckweed, brewers spent yeast and moringa leaves may limit their use for young growing fish. Overall, there is great potential for using several of the feed ingredients tested to replace fishmeal in diets for tilapia in Tanzania and across the East Africa region.

Fishmeal is considered to be the preferred source of dietary protein in commercial fish feeds [10,26]. However, the availability is declining and the price of fishmeal is increasing due to the declining global supplies of wild fish and competition with humans and other animals as a source of food or feed [10,27,28]. Moreover, the replacement of fishmeal with plant protein sources for cultured fish could be a considerable economic advantage due to lower feed costs [8,10,29,30].

With the exception of duckweed (AD, 46%), all test ingredients showed a high AD of protein (>77%). A high AD of protein in tilapia fed feed ingredients of varying origin has also been reported by others [9,20,31–34]. The duckweed used in the present study had a lower protein content and a higher fiber content than previously reported [35–37], which may explain the low AD of protein. The protein content of duckweed is reported to vary widely depending on plant age, environmental temperature and nutrient content of the aqueous environment [35,36]. Our results indicated that freshwater shrimp, marine shrimp, cattle blood, fish frames, brewers spent yeast and moringa leaves have acceptable protein digestibility to be used in tilapia diet formulation. The AD of protein in freshwater shrimp exceeding 100% can be explained by the low inclusion level (30%) of the ingredient in the test diet and the calculation of the AD value using the by-difference method [6].

The AD of energy was low in brewers spent yeast and duckweed (14–20%) with higher values in moringa leaves (32%), marine shrimp (62%) and fish frames (77%). The highest AD of energy was found in cattle blood and freshwater shrimp. The AD of energy for moringa leaves was similar to data for a range of plant feed ingredients evaluated in tilapia [29] while the AD values for brewers spent yeast and duckweed were lower (14–20%). Plant feed ingredients and plant by-products with a high fiber content usually give a lower AD of energy in tilapia [9,29,30]. A high AD of energy in fishmeal in tilapia was reported [9,18,31] while crayfish exoskeleton meal and gammarid meal showed lower AD values [20]. The low energy digestibility for brewers spent yeast and moringa leaves may limit their use as feed for young growing fish.

The AD of dry matter showed a large variation between test ingredients but was within the range of values reported for plant and animal ingredients in tilapia [10,20,33]. The variation can be explained by different fiber and chitin contents and the presence of other anti-nutritional compounds [6,38–40].

4. Materials and Methods

4.1. Study Site

The experiment was conducted at the Institute of Marine Sciences Mariculture Centre (IMS-MC) in Pangani, in the Tanga region of Tanzania (05°25′54.80″ S; 038°57′28.87″ E). The climate in Pangani is tropical with a mean annual temperature of 27 °C and a mean annual rainfall of 1214 mm.

4.2. Experimental Design

Eight protein source ingredients were selected for assessment of digestibility using siphoning or stripping for faeces collection.

Around 500 Nile tilapia fish with an average body weight (BW) 23.6 ± 0.3 g were purchased from a local tilapia fish farmer in Pangani. The fish were acclimatized for two weeks in 15 plastic tanks (1000 L) while being fed a locally made diet composed of fishmeal as the primary protein source (32% CP DM). After acclimatization, 480 healthy fish were selected and distributed into 48 plastic tanks (1000 L) with 10 fish per tank.

The tanks were divided into eight groups with six tanks per group. One group was assigned for the reference diet (control) and seven groups received test diets. The faeces collection method was applied in a change-over design with three tanks in each treatment group assigned for siphoning and the other three tanks assigned for stripping in the first experimental period, but swapped over to the other method in the second experimental period. The total experimental period was 56 days with 28 days per period.

The eight experimental diets comprised a reference diet with fishmeal as the main protein source and seven test diets containing 70% of the reference diet and 30% test protein ingredients (i.e., 70:30) on a dry weight basis. The experimental diets were formulated as suggested by Cho and Slinger [41]. Prior to the start of feeding the experimental diets, the fish were starved for four days to enhance the fish appetite during the feeding trial. Prior to handling, the fish were anaesthetized with clove

oil (100 mg L^{-1}). The present study was carried out in accordance with the law on the protection of animals against cruelty (Act no. 12/1974. of the United Republic of Tanzania).

4.3. Experimental Diets and Feeding

The selected feed ingredients were Lake Victoria sardines (*Rastrineobola argentea*; fishmeal, FM) (reference) and brewers spent yeast (BSY), moringa leaves (*Moringa oleifera*; ML), freshwater shrimp (*Caridina nilotica*; FSH), marine shrimp (*Exhippolysmata oplophoroides*; MSH), cattle blood (CB), duckweed (*Lemna minor*; DW) and fish frames (Nile perch, *Lates niloticus* skeletal remains; FF) (Table 1). Other ingredients included to balance the nutrient content in the diets were maize bran (MB), cotton seed cake (CSC), wheat pollard (WP), sunflower seed cake (SFSC), cassava flour, sunflower seed oil and a mineral and vitamin premix (Table 2). Approximately 0.5% (dry weight) of chromic oxide (Cr_2O_3) was included in the diets as an indigestible marker for the assessment of digestibility. The fish subjected to siphoning were fed once daily in the morning (09:00), at 4% of BW, which corresponded with feeding to satiation. The fish subjected to stripping were fed twice per day in the morning (09:00–10:00) and in the evening (15:00–16:00).

4.4. Faeces Collection

Siphoning: faeces were collected directly from the experimental tanks by siphoning [42]. In brief, feed wastes were removed daily through siphoning 2 h post feeding (11:00–12:00). Thereafter, faeces collection started and continued during the night. The faeces voided after the feed waste removal were collected in the afternoon (14:00–17:00) and in the morning of the following day (7:00–8:00). The siphoned faeces were collected on a 100 µm nylon filter mesh before being placed into appropriate sample collection containers. The process was repeated daily throughout the experimental trial. The daily fecal matter samples collected from each tank during the experiment were pooled within a tank and then kept frozen at −20 °C.

Stripping: faeces were collected (stripped) by pressing the belly to cause faeces to be expelled from the gut of each experimental fish [43]. The experimental fish were stripped into a sample collection container twice a week. The procedure was repeated throughout the experimental period. The faeces samples collected from the fish in each tank were pooled within a tank and kept frozen at −20 °C.

At the end of the experimental trial, all frozen fecal samples were thawed, mixed within the tank and oven-dried at 60 °C for 72 h and then stored at 4 °C for further analysis.

4.5. Water Quality Measurements

The water quality parameters were monitored weekly. The salinity was measured using a refractometer (RHS-10ATC, Shenzhen, China) and averaged 3.72 ± 0.70 ppt. The temperature and dissolved oxygen were measured by using a DO meter (HI-8424N, 161 Kallang Way, Singapore) with an average DO and temperature of 6.25 ± 1.04 mg/L and 23.9 ± 0.3 °C, respectively.

4.6. Calculations of Digestibility

The apparent digestibility (AD) of dry matter (AD_DM), organic matter (AD_OM), crude protein (AD_CP) and gross energy (AD_GE) in test and reference diets was determined according to Cho and Kaushik [44] as follows:

$$\text{AD\_DM (\%)} = 100 \times (1 - [\%\ \text{Dietary chromic oxide}/\%\ \text{Fecal chromic oxide}]) \quad (1)$$

$$\text{AD\_Nutrient (or Energy) (\%)} = 100 \times (1 - [\%\ \text{Fecal nutrient}/\%\ \text{Dietary nutrient}] \times [\%\ \text{Dietary chromic oxide}/\%\ \text{Fecal chromic oxide}]) \quad (2)$$

Cr_2O_3 values determined for faeces samples were normalized between sampling periods.

The AD of test ingredients (ADtest ingr.) was calculated as [45]:

$$\text{AD test ingr. (\%)} = \text{AD test diet} + ([\text{AD test diet} - \text{AD ref. diet}] \times [0.7 \times \text{D Ref}/0.3 \times \text{D test ingr.}]) \quad (3)$$

where D ref and D test ingr. are percentages of nutrients in the reference (fishmeal) and in the test ingredients, respectively.

4.7. Chemical Analysis

A proximate analysis of faeces and feed samples was performed at Sokoine University, Morogoro, Tanzania according to Association of Official Analytical Chemists (AOAC) [46]. In brief, dry matter (DM) was determined by drying 2 g of the sample (E 115, WTB binder 7200, Tuttlingen, Germany) at 105 °C overnight to a constant weight. The crude protein (CP) content was analyzed according to Kjeldahl [47] using a 2200 Kjeltec auto distillation unit (Foss, Tecator, Sweden). The crude fat (expressed as ether extract, EE) content was determined with petroleum ether (ST 243 SoxtecTM, Hilleroed, Denmark). The crude fiber (CF) was determined using a fiber analyzer (ANKOM 200 Fiber Analyzer, New York, NY, USA). The ash content was determined as the residue remaining after the incineration of 1 g of the sample in a muffle furnace at 550 °C for 3 h [48]. The nitrogen-free extract (Nfe) was calculated by subtracting the sum of moisture, CP, EE, CF and ash from 100 [49]. The gross energy (GE) in feed and faeces was calculated as 5.72*CP + 9.50*EE + 4.79*CF + 4.17*Nfe from the analyzed content (g kg^{-1} DM) of CP, EE, CF and Nfe [50].

The chromic oxide content in faeces and feed samples was analyzed in the Chemistry Laboratory at the University of Dar es Salaam by atomic absorption spectrometry (Varian AAS 240, Santa Clara, CA, USA) according to Hill et al. [51]. The samples were subjected to the wet acid digestion procedure according to Fenton and Fenton [52] with little modification. In brief, 250 mg of the sample was weighed into a digestion tube (100 mL) followed by an addition of 10 mL concentrated sulphuric acid (H_2SO_4) and 1 mL perchloric acid ($HClO_4$). The mixture was digested for 10–20 min until the contents turned yellow. The digested samples were then cooled and diluted to 50 mL with distilled water followed by a 20-fold dilution prior to a chromium content determination. The dilutes were analyzed for chromium (Cr) content by atomic absorption spectrometry (Varian AAS 240, Santa Clara, CA, USA) with an absorbance reading at 357.9 nm. The standard stock solutions used were 2, 5 and 10 mg/L of chromium (III) oxide.

4.8. Statistical Analysis

The AD values for DM, OM, CP and energy were subjected to analysis of variance (ANOVA) within and between faeces collection methods using SAS software (SAS version 9.4). The faeces collection method was considered a fixed effect in ANOVA while the diet was considered a random effect. The differences in AD values between diets were determined by pair-wise using the Tukey test and were considered significant at $p < 0.05$.

5. Conclusions

The dietary protein digestibility data obtained here suggest that with exception of duckweed, all other feed ingredients tested can be used to replace fishmeal without any reduction in protein value. With the exception of brewers spent yeast and duckweed, the test ingredients, particularly cattle blood, freshwater shrimp, fish frames and marine shrimp, can also support high dietary energy digestibility. Thus, several of these feed ingredients have the potential to be a replacement for fishmeal in diets for tilapia in Tanzania. This would contribute to a more long-term sustainable domestic fish production.

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