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Availability and Nutritive Value of *Spirulina* (*Arthrospira fusiformis*) from Arenguade and Chitu Lakes of Rift Valley of Ethiopia and Farmers' Perception about Its Utilization

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Abstract

Farmers perception about availability and utilization of Spirulina (*Arthrospira fusiformis*) were assessed and its nutritive value compared against reference proteins (soybean and nuge cake). Samples of Spirulina were collected from Arenguade and Chitu lakes of rift valley of Ethiopia. A survey was conducted on purposively selected 100 households (HH) living around the Lakes which are located in Gerbicha and Labu subuqa districts of Debre Zeit town and west Arsi zone, respectively. Chemical composition and *in vitro* dry matter digestibility (IVDMD) and total gas volume (TGV) of feed samples were determined. Respondents of both districts are not aware of Spirulina as animal feed. Spirulina was available during dry season in Arenguade and whole year in Chitu. The highest ($p < 0.05$) ash (26%), ether extract (EE, 13%), crude protein (CP, 33%) and IVDMD (96%) was recorded for Spirulina from Lake Arenguade. There was no significant difference ($p > 0.05$) in crude fiber (CF), neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL) and metabolizable energy (ME) contents between Spirulina from both lakes. Spirulina from Chitu produced the highest TGV (31.2 ml) and fermented faster than Spirulina from Arenguade. Lowest ($p < 0.05$) ash, EE, CP and IVDMD and higher ($p < 0.05$) CF, NDF, ADF, ADL and TGV were recorded for reference proteins than Spirulina. ME content of Spirulina (3200 kcal/kg) was higher ($p < 0.05$) than that of nuge cake (3011 kcal/kg) but lower ($p < 0.05$) than that of soybean (3474 kcal/kg). Therefore, the chemical composition and IVDMD showed the potential of Spirulina to be used as a substitute of the reference feeds.

Keywords

Spirulina, Lake Arenguade, Lake Chitu, Reference Feed, Chemical Composition

1. Introduction

Livestock are important component of nearly all farming systems in Ethiopia and serve as source of draught power, milk, meat, manure, hides, skins and cash income. In spite of this, the productivity of livestock is low due to production constraints, socio-economic and technical limitations [1]. Inadequate feed, widespread diseases, poor breeding stock, inadequate credit, extension, marketing and infrastructure are the major constraints affecting livestock performance in the country [2], poor nutrition being the most limiting factor [3]. It was also stated that the major constraint to livestock production in developing countries is the fluctuation in quantity and quality of feeds [4]. The same authors suggested that these countries experience serious shortages of the conventional feeds. With an increasing demand for livestock products as a result of rapid growth in the world economies and shrinking land area, future hopes of feeding the millions and safeguarding their food security will depend on the better utilization of non-conventional feed resources, which cannot be used as food for humans [4]. Livestock feed cost in developing countries is a challenge. The high and increasing prices for animal feeds have compelled researchers in these countries to direct their attention to non-conventional feeds, with particular emphasis on protein substitutes [5].

The productivity of livestock and poultry in the tropics has been grossly limited by the scarcity and consequent high prices of the protein and energy sources [6]. Although soybean meal and fish meal have been widely and successfully used as reference protein sources for livestock, their prices have been escalating, whilst availability is often erratic [7]. In many developing countries animal source feed ingredients are not preferred because of high price for small scale farmers, which is a compelling reason for search of potentially useful locally available, alternative feedstuffs. The identification of new feed resources is, therefore, crucial for sustainable animal production and future viability. Ideally, the new feed resource should have high nutritive value and conversion efficiency, be able to optimize animal product quality and use land and water efficiently [8].

Thus, *Spirulina* has received special attention, because, these microorganisms can be a good alternative source of protein in the diet [9]. Among several microorganisms which have been studied, the blue-green algae, *Spirulina,* is considered a promising microorganism due to its high protein (65% to 70% DM), vitamins and minerals [10] [11]. It is estimated that Spirulina can produce 125 times more protein than that of corn when grown on the same area of corn [12]. *Spirulina* fusiformis is a microalgae with appropriate composition to be used as a food supplement. It is commonly used by humans and animals as protein source. Several studies such as palatability, lack of toxicity and easy digestion, antioxidant actions, hypocholesterolemic, anticancer, immuno-stimulant, anti-inflammatory, antiviral, among others have been conducted to verify the possible benefits of *Spirulina* and some properties have been verified [13] [14] [15].

The addition of 1.5% to 12% of *Spirulina* in to the diets of broilers can replace other protein sources, especially soybean meal, with satisfactory growth rates and feed efficiency [16]. However, the optimal levels for using *Spirulina* as a substitute of conventional proteins in a diet are still debatable [17] [18] [19].

Algae contains proteins, carbohydrates, lipids and trace nutrients, including vitamins, antioxidants, and trace elements. The nutritional contents of algae are rapidly gaining importance as a renewable source to substitute the conventional ingredients in aquaculture/animal feed. Therefore, *Spirulina* is emerging as a cost effective means of improving animal productivity for a sustainable and viable food security [8].

Even though *Spirulina* occurs naturally in different rift valley lakes of Ethiopia there is limited information available with regard to its utilization and nutritive value. Thus this study was designed to assess availability and farmer's perception about utilization of *Spirulina* and compare its nutritive value (nutrient composition, *in vitro* dry matter degradability and gas production) against reference protein supplements (soybean and nuge cake).

2. Materials and Methods

2.1. Description of the Study Area

Lake Chitu and Arenguade were selected based on the availability of *Spirulina*. Lake Chitu (**Figure 1**) is a volcanic explosion crater lake which is located 287 km South of Addis Ababa in West Arsi zone, Aje district of Labu Subuqa district. Lake Chitu is situated at 25 km from Shashamane town with latitude 7˚24'26"N,

Figure 1. Lake Chitu.

longitude of 38˚25'33"E and an altitude of 1540 meters above sea level and an area of 0.8 km^2 and a maximum depth of 21 m. It is among lakes of the National Park of the Ethiopian Rift Valley.

Lake Arenguade (**Figure 2**) is a member of crater lakes –Bishoftu crater lakes– situated 47 km South East of Addis Ababa in Debre Zeit town of Gerbicha district at a latitude of 8˚41.856'N and 38˚58.796'E and altitude of about 1900 m, above sea level. Lake Arenguade is an alkaline soda lake with a diameter of approximately 800 m and maximum and mean depth of 32 and 18 m, respectively. Lake Arenguade is so named (Arenguade means green in Amharic language) after the green coloration of surface water by the filamentous blue-green algae.

2.2. Sample Collection Procedure

Samples of *Spirulina* were collected from Lake Arenguade and Lake Chitu from four different directions of each lake (north, south, east and west) during the months of October and April. *Spirulina* samples collected from different directions were used as replications for each lake. The collection was carried out by skimming the surface using a pot and draining/filtering it out. The water was let to pass through a sieve to control passage of dirt and then was further filtered by clean cloth to separate the water from *spirulina*. And then the sample was air dried for three days. The drying process was done inside the house (in a shade) on a concrete floor. The reference feed sources (nuge seed cake and soybean) were purchased from local market. Soybean was roasted for 5 - 10 min in the laboratory of Animal and Range Sciences of College of Agriculture at Hawassa University.

2.3. Survey

From household (HH) living in Gerbicha and Labusu buqa districts near the lake, 50 HH were selected purposively from each district around lake Arenguade

Figure 2. Lake Arenguade.

and Chitu respectively. Around each lake, livestock owners were briefed about the objective of the study before data collection. Information about availability, farmers' awareness about *Spirulina* and form of utilization in the area both by humans and animals were collected using structured questioners. The interview was conducted at the residences of livestock owners with the assistance of agricultural experts or development agents.

2.4. Chemical Analysis

Samples of *Spirulina,* nuge seed cake and soybean were ground to pass through 1 mm sieve size. Dry matter percentage of the samples was determined by oven drying the samples at 105˚C for 24 hours. Total ash and CP contents were determined [20]. Ash was determined by complete burning of the feed samples in a muffle furnace at 500˚C overnight. Nitrogen was determined using the Kjeldhal procedure and crude protein was calculated as

$$\text{CP} = \text{N} \times 6.25 . \quad (1)$$

Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were analyzed using the detergent extraction method [21]. Metabolizable energy (ME) of the feed sample was calculated as:

$$\text{ME}\left(\text{Kcal/kg DM}\right) = 3951 + 54.4\text{EE} - 88.7\text{CF} - 40.8\text{Ash} \quad [22]. \quad (2)$$

2.5. *In Vitro* Dry Matter Digestibility

An *in vitro* dry matter digestibility (IVDMD) of *Spirulina*, nuge seed cake and soybean was determined by the method [23] which was later modified [24]. Rumen fluid was collected from three sheep which were kept at Hawassa University Animal and Range Sciences farm and were fed on medium quality diet (200 g concentrate and *ad-libitum* hay). The rumen fluid was collected by sucking using rumen tube before the morning feeding and placed directly into pre-warmed thermos flasks and taken immediately to the laboratory. It was then filtered through two layers of cheese cloth. About 0.5 g of the dried and ground samples from each feed was put in flasks containing rumen fluid-medium mixture and incubated for 48 hours at 39˚C. Microbial digestion was followed by neutral detergent refraction and the indigestible residue was measured after overnight oven drying at 105˚C.

2.6. *In Vitro* Gas Production

200 mg of each sample (ground to pass through a 1mm sieve) were put in to 100 ml calibrated glass syringes in three replicates, fitted with Vaseline lubricated pistons. To prepare the inoculum, rumen fluid was collected before the morning feeding from three sheep. The rumen fluid was placed directly into pre-warmed thermos flasks and taken immediately to the laboratory. It was then filtered through two layers of cheese cloth and diluted with buffered mineral solution as previously described [25], which was maintained in a water bath at 39˚C under

continuous flushing with CO_2. A total of 30 ml incubation medium was transferred into glass syringes containing the samples and three blank syringes. They were then immediately placed in a temperature controlled incubator preset at 39˚C. The initial volume of each syringe was recorded before the commencement of the incubation of the samples. The gas volume was recorded after 4, 8, 12, 24, 48, 72 and 96 hours of incubation.

The result was fitted in to the exponential equation:

$$p = a + b\left(1 - \mathrm{e}^{-ct}\right) \tag{3}$$

where p = gas production at time t, a = gas production from the immediately fermentable organic matter (the intercept of the gas production curve), b = gas production from fermentation of the slowly but potentially fermentable organic matter, $a + b$ = potential gas production (the asymptote of the gas production curve), c = rate of gas production, t=incubation time, and e=base of natural logarithm [26] [27]. Gas production data was estimated using Neway Excel program [28].

2.7. Statistical Analysis

All the statistical analyses were carried out using a Statistical Package for Social Science (SPSS, version 20). The survey data was analyzed using descriptive statistics. One way independent mean comparison of parameters was used to compare data between the two study sites. Means of chemical composition, IVDMD and metabolizable energy were separated using Tukey HSD (Tukey honestly significant difference) test. The model used for chemical composition and IVDMD was:

$$Y_{ij} = \mu + A_i + B_j + e_{ij} \tag{4}$$

where Y_{ij} = response variable, μ = overall mean, A_i = variation due to lake, B_j=variation due to sampling spot/site and e_{ij} = random error effect.

3. Results

3.1. General Characteristics of Sampled Households

The majority of respondents were male and large numbers of respondents were illiterate in both study areas and very few had educational background of basic education and primary school. The age of respondents in Lake Arenguade and Chitu was 37.62 ± 9.9 and 38.96 ± 8.6 years, respectively.

The average land holding of respondents in Lake Arenguade and Lake Chitu was 1.6 and 1.8 ha, respectively. The result shows that from the total land holding, most of it was used for crop cultivation in both study areas. Their major livelihood activity in the two study sites was mixed crop livestock farming. Crop production was major source of revenue in both study areas contributing to 32% of the livelihood of the respondents. The dominant crops produced are teff, maize, wheat and barley; and they are produced once in a year. Livestock contributes to 16% of the livelihood of the lake community and is used for agricultural activities, income generation and sustaining family protein needs. Cattle and goat were the dominant species of livestock in the study areas followed by donkey and the least were sheep. There was greater number of livestock per households around Lake Chitu than Arenguade. The main purpose of keeping different groups of livestock species was for traction (68% and 42%) followed by transportation (28% and 22%) as reported by respondents of Lake Arenguade and Lake Chitu, respectively. Few respondents keep livestock for milk and meat production, breeding and prestige.

3.2. Feed Resources Availability and Perception of Farmers about Use of *Spirulina*

As observed from the interviewees reply the availability of feed in the two study areas was similar. Crop residues such as teff straw, wheat straw and maize stover are the common and most important feed resources of livestock around both lakes followed by communal grazing land and crop aftermath. Some of the respondents supplement their livestock during feed shortage by purchasing supplementary feed. Majority of respondents indicated wet season as the season of higher feed shortage (50% and 42%) while less number of them said dry season (16% and 10% of respondents) and 34% and 48% of respondents said shortage of feed is independent of season in Lake Arenguade and Chitu, respectively.

Feed shortage was the major constraint identified by respondents of the two study sites. Respondents around Lake Arenguade and Lake Chitu alleviate feed shortage by conservation of feed in the form of hay [31 (62%) and 27 (54%)], supplementation [16 (32%) and 13 (26%)] and destocking [3 (6%) and 10 (20%)], respectively

The availability of *Spirulina* was season dependent in Lake Arenguade where 86% of the households reported that it is available only during dry season. In Lake Chitu the majority of the respondents (92%) stated that it is available year-round even though its biomass fluctuates between dry and wet seasons.

Spirulina was not used in both study areas as animal feed, but some of the respondents (42%) in Lake Arenguade reported that livestock owners who live in and around Debre Zeit bring their cattle to the lake to drink the water considering it has medicinal value for animals. Almost all respondents from Lake Arenguade and Chitu indicated that cattle do not consume *Spirulina*. They only drink the water by removing the floating *Spirulina* bloom by their breath.

As reported by farmers flamingoes in Lake Arengude are rarely available and 88% of the respondents said that they had no idea if flamingoes feed on *Spirulina*, while the remaining 6% of respondents said that they feed on *Spirulina* and others (6%) said they feed on aquatic animals. All farmers around Lake Chitu indicated that flamingoes are there the whole year in a huge number. About 52% of respondents around Lake Chitu alleged that the reason for a huge number of flamingoes in the lake was to feed on *Spirulina* while 22% of the respondents said that they feed on aquatic animals and 26% of respondents do not have any idea about what flamingoes feed on.

3.3. Chemical Composition, *In Vitro* Digestibility and Gas Production Traits

Chemical composition and nutritive value of *Spirulina* from Lake Chitu and Arengude and the referenced feeds (roasted soybean and nuge cake) are shown in Table 1.

The ash content differed significantly ($p < 0.05$) between *Spirulina* samples and much more significantly between *Spirulina* and the reference feeds, *Spirulina* having much higher ash content than the reference feeds. There was no significant difference ($p > 0.05$) in the crude fiber content of *Spirulina* from the two lakes but their crude fiber content differs significantly ($p < 0.05$) from the reference feeds. The ether extract content of *Spirulina* was significantly ($p < 0.05$) influenced by the sampling site. As shown in Table 1 the ether extract content of *Spirulina* was higher than ($p < 0.05$) the reference feeds. The NDF and ADF content of *Spirulina* from the two lakes and that of soybean did not differ ($p > 0.05$) but NDF and ADF content of nuge cake was higher than that of *Spirulina* from both lakes and soybean. There was no significant ($p > 0.05$) difference in ADL value between *Spirulina* of the two Lakes. The ADL value of the reference feeds (roasted soybean and nuge cake) was higher ($p < 0.05$) than that of *Spirulina*, nuge cake having the highest value. The CP content of *Spirulina* from Lake Arenguade was higher than that of Lake Chitu. The CP content of *Spirulina* was higher ($p < 0.05$) than the reference feeds. *Spirulina* from Lake Arenguade had the highest ($p < 0.05$) IVDMD followed by *Spirulina* from lake Chitu.

Pattern of gas production of *Spirulina* samples from Lake Arenguade and Chitu and the referenced feeds is given in Figure 3.

Table 1. Chemical composition and *in vitro* dry matter digestibility of *Spirulina* from Lake Arenguade and Chitu and reference feeds.

Parameters (% DM unless specified)	Spirulina		Reference feeds	
	Lake Arenguade	Lake Chitu	Soybean	Nuge cake
Dry matter (%)	96.42 ± 0.06^b	96.44 ± 0.06^b	98.46 ± 0.13^a	98.66 ± 0.13^a
Ash	26.18 ± 0.08^a	23.03 ± 0.08^b	8.6 ± 0.16^d	12.39 ± 0.16^c
Crude fiber	4.60 ± 0.14^c	4.20 ± 0.14^c	6.80 ± 0.28^b	11.50 ± 0.28^a
Ether extract	13.30 ± 0.08^a	11.80 ± 0.08^b	8.80 ± 0.17^d	10.80 ± 0.17^c
Neutral detergent fiber	19.60 ± 0.37^b	18.80 ± 0.37^b	19.90 ± 0.74^b	39.17 ± 0.74^a
Acid detergent fiber	7.02 ± 1.01^b	6.40 ± 1.01^b	12.60 ± 2.03^b	23.80 ± 2.03^a
Acid detergent lignin	1.80 ± 0.14^c	1.58 ± 0.14^c	4.08 ± 0.28^b	13.30 ± 0.28^a
Crude protein	33.30 ± 0.05^a	31.30 ± 0.05^b	29.60 ± 0.01^c	22.20 ± 0.01^d
ME (kcal/kg)	3203 ± 12.6^b	3281 ± 12.6^b	3474 ± 25.18^a	3011 ± 25.18^c
IVDMD (%)	96.67 ± 0.18^a	95.34 ± 0.18^b	92.67 ± 0.36^c	65.43 ± 0.36^d

ME = metabolizable energy; IVDMD = *in vitro* dry mater digestibility; means with different superscript letter across row are significantly different ($p < 0.05$).

Nuge cake fermented faster and *Spirulina* sample from Lake Arenguade ferments slower than soybean and *Spirulina* sample from Lake Chitu. There was a steady increase in gas production after 24 hr incubation time. The cumulative gas production increased during the incubation period.

As shown in Table 2 *Spirulina* from Lake Chitu was fermented faster (C) than all other feeds and it was followed by roasted soybean. The highest intercept of the gas production curve (A) was in reference feeds; and the lowest in *Spirulina*. *Spirulina* from Lake Chitu had negative "A" values. Gas production from the slowly fermentable dry matter (B) was the highest in *Spirulina* from Lake Chitu and the lowest in *Spirulina* from Lake Arenguade. The potential gas production (a+b) was higher for the reference feeds than *Spirulina* of the two lakes; and *Spirulina* from Lake Chitu had higher potential gas production than that of Lake Arenguade.

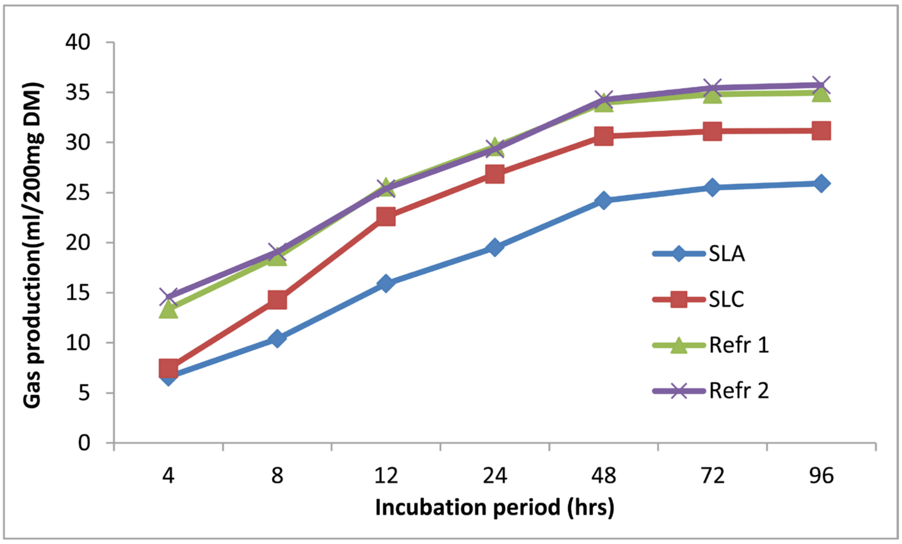

Figure 3. The pattern of fermentation of *Spirulina* was distinctly different from the referenced feed during the incubation time. SLA = *Spirulina* sample from Lake Arenguadea; SLC = *Spirulina* sample from Lake Chitu; Refr 1 = roasted soybean and Refr 2 = Nuge cake.

Table 2. Gas production characteristics of *Spirulina* from Lake Chitu and Arenguade and reference feeds.

Estimated parameter	Feedstuffs			
	Spirulina LA	*Spirulina* LC	Soybean	Nuge cake
C	0.055	0.085	0.069	0.059
A	1.9	−2.1	6.5	8.9
B	24.1	33.3	28.5	26.9
a + b	26.0	31.2	35.0	35.8

LA = Lake Arenguade; LC = Lake Chitu; A = the gas production from the immediately soluble fraction; B = the gas production from the insoluble fraction; C = the gas production rate constant for the insoluble fraction b (h); a + b = potential gas production.

4. Discussion

The average land holding in both the study areas is lower than those reported for other studies [29] [30] possibly due to differences in population density of the areas. The livelihood of respondents in both the study areas is mainly based on agriculture and it is of mixed crop livestock production system. This result is in line with earlier reports that mixed agriculture is the major livelihood activity (72.5%) of people living around Chitu Lake and the contribution of livestock in supporting the livelihood of lake community is of paramount importance [31]. Respondents of both lake communities keep mixture of livestock species for different reasons [32]. Feed shortage was the major livestock production constraint in both the study areas, crop residues being the main feed resource throughout the year. This result agrees with earlier report that natural pasture and crop residues are the major feed resources of Ethiopia [33]. The livestock production constraints reported in this study are in agreement with the observations that inadequate feed and nutrition, poor health, low productivity of local breeding stock are the main livestock production constraints in Ethiopia [2].

Communities around both lakes were not feeding *Spirulina* to their animals even though it is available in both lakes. Respondents from around Lake Arenguade believe that lake water has medicinal value for animals which agrees with earlier report where local people were encouraging their cattle to drink lake water believing that the *Spirulina* water has some therapeutic effects and compensates for some deficiency in diets of cows [34] [35].

The ether extract (EE) content of *Spirulina* was higher than that of the reference feeds (11% - 13%). It has been reported earlier that the EE content of *Spirulina* ranges from 5.6% to 7% [36] [37] and from 6% to 13% [38] [39]. As can be seen most of EE values earlier reported are lower than the findings in this study. The crude fiber content of *Spirulina* from both lakes was lower than that of the reference feeds. It was revealed that the fiber content in *Spirulina* varied from 4% to 7%; the results of the present study fall within this range, but very close to the lower range [40]. However, crude fiber content in *S. platensis* was reported as 0.5% which is extremely lower than the values obtained in this study [41]; and a fiber content of *Spirulina* ranging from 8% to 10% was also reported which is again much higher than the values in this study [42]. The ash content of *Spirulina* in this study was higher than that of the reference feeds. The ash content from the current study was much higher than the values (6.4% - 9%, 7% and 8.41%) earlier reported [37] [43] [44] [45]; respectively. This might be due to the difference in geographical location of the sample. Many factors may affect the bioaccumulation of minerals in *Spirulina* and the most important ones were differences in culture media, temperatures, pH and salinity [42]. Crude protein content of *Spirulina* was reported as 58.2% [46]. Also higher CP content (69.2%) was also obtained [45] in *Spirulina* from Kanem Lake Chad. The differences could be explained by differences in either the environment, or climate, or the techniques used to collect *S. platensis* [47]. Metabolizable energy content of Spirulina in the current study is similar with literature results (2500 to 3290 kcal/kg) reported [48] [49] [50].

The IVDMD of *Spirulina* was higher than the reference feeds which may be due to the higher level of fiber in the reference feeds when compared to *Spirulina* which agrees with earlier results reported and the decrease in IVDMD may be associated with its high lignin content as feeds with less lignin promote better access to microorganisms [51]. Furthermore, *Spirulina* cell walls are mucopolysaccharides (complex sugars) that are easily digestible. Digestibility tests have shown *Spirulina* to be 83% to 95% digestible [52].

In the current study, the gas production of *Spirulina* was lower than the reference feeds throughout the incubation period which may be due to higher CP content of *Spirulina*. Even though gas production (A and B) of the reference feeds was higher than *Spirulina* the potential gas production of the reference feed was lower. This result is in line with earlier reports that stated the potential of gas production (a + b) for protein feed source was lower than that of carbonaceous concentrate feedstuffs [53]. It was also reported that protein fermentation does not lead to extensive gas production [54]. Besides, the lower gas volume of *Spirulina* might be further clarified by the presence of high fat, which contributes to negligible gas production [55].

5. Conclusion

Spirulina samples collected from Lake Chitu and Lake Arenguade differ in their ash, ether extract, crude protein, IVDMD and TGV. *Spirulina* has a comparable DM and ME but lower fiber and TGV and higher ash, EE, CP and IVDMD than the reference feeds (soybean and nuge cake). *Spirulina* is thus better than the reference feeds and can be used as a substitute for the reference protein supplements in animal feed because it is available most of the time in both lakes and can also minimize feed cost. Results of this study indicated feed shortage as the major livestock production constraint in both study areas and crop residues as the major feed resources available are low in quality to meet the requirement of livestock and thus Spirulina can partly offset the shortage. Respondents of both lake communities are not feeding *Spirulina* to animals; therefore, the use of *Spirulina* in animal feed should be encouraged through awareness creation to improve livestock production and productivity in the study areas. Further researches of the effect of feeding *Spirulina* on animal performance are suggested.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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