



## Effects of processing on Duckweed (*Lemna minor*) as fish feedstuff

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### Abstract

The nutritional value of duckweed (*Lemna minor*) has been investigated as a suitable substitute for soybean meal in aquafeed. However, information on its processed forms in the diets for better nutrients utilization are yet to be reported. Therefore, this research investigated the effects of treated duckweed meal (DWM) as soybean substitute. The proximate composition, anti-nutritional factors and amino acids concentration of raw (Ra), cooked (Co), and soaked in potash (So) DWM were evaluated using standard methods. The crude protein of the test ingredients varied from 22.70% (Ra) to 29.45% (Co). Anti-nutrient factors of saponin, tannins, and cyanide were significantly ( $p < 0.05$ ) reduced from  $0.72 \pm 0.01\%$  to  $0.41 \pm 0.01\%$ ,  $64.00 \pm 0.29$  (mg/100g) to  $20.50 \pm 0.29$  (mg/100g) and  $4.88 \pm 0.01$  (mg/100g) to  $2.61 \pm 0.35$  (mg/100g) in Ra and Co samples, while oxalate and phytate significantly ( $p < 0.05$ ) reduced from  $219.00 \pm 0.58$  to  $65.00 \pm 0.58$  (mg/100g) and  $732.84 \pm 0.37$  to  $165.80 \pm 0.52$  (mg/100g) in Ra and So, respectively. However, the value of alkaloid (%) was significantly ( $p < 0.05$ ) increased from  $4.10 \pm 0.58$  (Ra) to  $4.70 \pm 0.01$  (So). There was also an increase in the total amino acids, total essential amino acids, and total non-essential amino acids of the treated (Co and So) DWM over the Ra, as well as the essential amino acid score of So over Co and Ra DWM samples. The implication of the processing revealed that



nutritional and amino acid content was enhanced with a substantial reduction in phytochemicals except for alkaloids.

**Keywords:** Anti-nutritional factors, Amino acid, Aquatic macrophyte, *Lemna minor*, Duckweed meal

## Introduction

Aquatic macrophytes availability has constituted a nuisance (Aghoghovwia et al., 2018) to the ecology of various water bodies due to high regenerative capacity within a short duration (Skillicorn and Williams, 1993; Leng et al., 1995). Most of these macrophytes have been assessed for their nutritional content (Sonta et al., 2019), anti-nutritional factors (Xu et al., 2021), and amino acid profile (Chakrabarti et al., 2018) using different processing methods (Sogbesan et al., 2015; Ifie et al., 2020) to increase the compositional value (Iskander et al., 2019; Irabor et al., 2022) of the macrophytes. Plant based feed ingredients are known to possess concentrated forms of anti-nutritional factors (ANF) locked up in the plant cell wall (Agbo et al., 2011), which affects the utilization by interacting with the intestinal tract of animals with effect on digestibility and absorption of protein and amino acid (Nowacki, 1980).

Duckweeds belong to the family *Lemnaceae* (Acosta et al., 2021; Liu et al., 2021) which represent a small family of aquatic floating monocots plant consisting of 37 species in 5 genera. It has global distribution and inhabits small shallow bodies of water in tropical and boreal environments (Acosta et al., 2021). In the angiosperm plants family, they are the fastest to grow (Sree et al., 2015); and form a continuous layer of mats on or underneath the water surface (Sonta et al., 2019). Stewart et al., (2021) noted that duckweed has the ability to convert wastewater into high-quality protein and grow under varied environmental conditions; using the nitrogen in the waste and its ammonium ion for optimal growth (Sonta et al., 2019). The environmental requirement of pH 5-9, temperature 6°C-33°C, with a growth life span of 3-10 weeks allows duckweed to rapidly regenerate in nutrient rich wastewater with adequate sunlight within this period before die-off (Liu et al., 2021).

The Suitability of *L. minor* over other species has been recognized due to its high dietary fiber content which enhances the ease of food digestibility in the digestive system of humans (Yahaya et al., 2022). Protein from plant sources can also be derived from aquatic plants which can be used as alternative feed ingredients (Andriani et al., 2019). Olasunkanmi et al., (2020) noted that aquatic plants are underutilized as feed ingredients, however, they can serve as alternatives to the expensive conventional feedstuff in the local and international human and animal nutrition

industry. Duckweed utilization in aquaculture is of importance in water quality management and as a feed ingredient source. The high nutritional content in terms of amino acid carbohydrates and protein makes it important (Said *et al.*, 2022), while the mechanism for the reduction of ANF in plant-based feed ingredients needs to be further studied to know their impact on cultured livestock (Naseem *et al.*, 2020). It has been noted that increased utilization of DWM can be achieved with processing, so as to control detrimental effects associated with DWM from bioremediation culture media (Ifie *et al.*, 2020). Elucidating these nutritional chemical constituents will be beneficial to the enhancement of the usage in various areas of concern (Xu *et al.*, 2021).

Duckweed has found usefulness in wastewater treatment (Singh *et al.*, 2012; Liu *et al.*, 2021), food preservative against fungal spoilage (Effiong and Sanni, 2009), and production of bio-ethanol (Muradov *et al.*, 2014). In recent times it has become a potential asset to farmers and animal nutritionists as feedstuff to the livestock industry as a soybean substitute for poultry (Ghosh *et al.*, 2015; Shammout and Zakaria 2015), catfish (Da *et al.*, 2013), carp and rohu (Sharma *et al.*, 2016). Sonta *et al.*, (2019) opined that collection and processing will further improve the utilization of DWM as a plant protein source for livestock and aquaculture. The objective of this study determined the effect of processing methods on the nutritional content of raw and treated duckweed meals.

## **Material and methods**

### **Duckweed sample collection**

About 125kg of fresh duckweed (*Lemna minor*) was harvested from the water surface on a fish pond. The weed was collected from the water surface using a large plastic sieve, drained, packed in draining sacks, and transported to the Wet Research Laboratory of the Department of Fisheries Technology, Federal College of Animal Health and Production Technology, Ibadan. The weight of the wet duckweed was taken using a 100kg capacity Camry top loading balance.

### **Duckweed preparation**

The fresh duckweed collected was divided into three (3) processing groups for preparation. Raw Duckweed: 37.5kg was air-dried under shade. Cooked Duckweed: 37.5kg was boiled at 100°C for 5 minutes (Sogbesan *et al.*, 2015), drained, and air-dried under shade. Potash Treated Duckweed: 37.5kg was soaked in potash (Maize cob ash) solution at 5g per liter for 24 hours (Vadivel and Pugalenth, 2008), drained and air-dried under shade (Ifie *et al.*, 2020). The dried product of the



processed duckweeds was milled into powder using home milling/grinding machine and packed in airtight polythene bag in preparation for feed production.

### Chemical analysis of experimental materials

Chemical analysis for proximate, anti-nutritional factors (ANF) and amino acid profiles of raw and processed duckweed samples was carried out. The proximate analyses were according to AOAC (2005) methods. The saponin and alkaloid content of the samples was determined by the double extraction gravimetric method as described by Harborne (1973). Phytate content was determined through phytic acid determination using the procedures of Lucas and Markaka (1975). Tannin was determined by using a slightly modified method of AOAC (1980), while total oxalate was determined by the modified method of Abeza *et al.*, (1968). The amino acids were quantitatively measured by the procedure of Spackma *et al.*, (1958) using an automated amino acid analyzer (Technicon Sequential Multi-sample Analyzer, TSM). The Predicted Protein Efficiency Ratio (P-PER) of the fresh sample was calculated from their amino acid composition based on the equation developed by Alsmayer *et al.*, (1974)  $P\text{-PER} = 0.468 + 0.454 (\text{Leu}) - 0.105 (\text{Tyr})$ .

### Statistical Analysis

Data obtained from this experiment were subjected to a one-way analysis of variance (ANOVA). Duncan's multiple range test was used to test differences among means when significant values are observed at  $P < 0.05$  using IBM SPSS version 21.

## Results

### Chemical characterization of raw and treated duckweed (*Lemna minor*) meals

The results of the proximate analysis of raw and processed duckweed meal (Table 1) revealed that CP and CF of treated samples were significantly increased ( $p < 0.05$ ) by processing over raw, while the ash content in treated samples was reduced but not significantly different ( $p > 0.05$ ) among the treated samples. The treatment methods tend to increase protein and fiber contents significantly ( $p < 0.05$ ) above that of the Ra, while lowering the ash and carbohydrates (NFE) in the samples.

**Table 1:** Proximate composition of raw and treated duckweed meal

Parameters (%)	Samples
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	Ra	Co	So
Dry Matter	92.02 <sup>a</sup>	92.40 <sup>a</sup>	91.95 <sup>ab</sup>
Moisture content	7.98 <sup>b</sup>	7.60 <sup>c</sup>	8.05 <sup>a</sup>
Crude Protein (CP)	22.70 <sup>c</sup>	29.45 <sup>a</sup>	25.50 <sup>b</sup>
Ash	19.80 <sup>a</sup>	17.50 <sup>b</sup>	17.00 <sup>b</sup>
Ether Extract	3.30 <sup>b</sup>	3.20 <sup>c</sup>	3.50 <sup>a</sup>
Crude Fibre (CF)	7.70 <sup>b</sup>	8.30 <sup>a</sup>	8.10 <sup>a</sup>
NFE	38.52 <sup>a</sup>	33.95 <sup>c</sup>	37.85 <sup>b</sup>

Rows Means ±SE with different superscripts are significantly different (p<0.05)

NFE = Nitrogen Free Extract, Ra = Raw duckweed meal, Co = Cooked duckweed meal, So = Soaked in potash duckweed meal

### Anti-nutritional factors of raw and treated duckweed meals (*Lemna minor*)

The anti-nutritional factors (Table 2) were analyzed in the samples composed of oxalate, tannins, cyanide, phytate, saponins, and alkaloids. It was observed that methods of processing significantly (p<0.05) lowered ANF of processed duckweed samples, while alkaloids were increased over the Ra with a significant (p<0.05) value. The significantly (p<0.05) high values of oxalate, saponin, tannins, cyanide, and phytate were recorded in Ra. All the parameters were significantly (p<0.05) different from each other among the raw and processing methods.

**Table 2:** Anti-nutritional factors of raw and treated duckweed meals (*Lemna minor*)

*ANF	Processing Methods		
	Ra	Co	So
Oxalate (mg/100g)	219.00±0.58 <sup>a</sup>	98.50±0.29 <sup>b</sup>	65.00±0.58 <sup>c</sup>
Saponins (%)	0.72±0.01 <sup>a</sup>	0.41±0.01 <sup>c</sup>	0.63±0.01 <sup>b</sup>
Tannins (mg/100g)	64.00±0.29 <sup>a</sup>	20.50±0.29 <sup>c</sup>	47.75±0.14 <sup>b</sup>
Cyanide (mg/100g)	4.88±0.01 <sup>a</sup>	2.61±0.35 <sup>c</sup>	4.47±0.02 <sup>b</sup>
Phytates (mg/100g)	732.84±0.37 <sup>a</sup>	216.17±0.31 <sup>b</sup>	165.80±0.52 <sup>c</sup>
Alkaloid (%)	4.10±0.58 <sup>c</sup>	4.38±0.01 <sup>b</sup>	4.70±0.01 <sup>a</sup>

Rows Means ±SE with different superscripts are significantly different (p<0.05)

\*ANF = Anti-Nutritional Factors, Ra = Raw duckweed meal, Co = Cooked duckweed meal, So = Soaked in potash duckweed meal

### Amino acids composition of raw and treated duckweed meals

The amino acids profile of raw and treated duckweed meal (Table 3) showed leucine as the most concentrated essential amino acid (EAA) (g/100g protein) and tryptophan as the least concentrated.

Glutamic acid was the highest concentrated non-essential amino acid (NEAA) and Aspartic acid was rated second most concentrated. Generally, Glutamic acid is the most concentrated amino acid in the raw and treated samples while cysteine is the least concentrated amino acid. The calculated predicted protein efficiency ratio from the amino acids was least in Ra and high in Co and So respectively.

**Table 3:** Mean Amino acids composition of raw and treated duckweed meals

<b>Amino Acids</b> (g/100g crude protein)	<b>Ra</b>	<b>Co</b>	<b>So</b>
Leucine (Leu)*	6.48	6.59	7.00
Lysine (Lys)*	5.04	5.35	4.80
Isoleucine (Ile)*	4.03	4.20	4.82
Phenylalanine (Phe)*	4.61	4.61	5.11
Tryptophan (Try)*	1.11	1.13	1.33
Valine (Val)*	4.56	4.59	5.01
Methionine (Met)*	1.92	1.22	2.25
Proline (pro)	2.81	2.96	3.01
Arginine (Arg)*	6.41	6.49	7.02
Tyrosine (Tyr)	3.22	3.01	3.66
Histidine (His)*	2.26	2.30	2.33
Cystine (Cys)	0.71	0.76	0.85
Alanine (Ala)	4.21	4.31	4.60
Glutamic acid (Glu)	11.16	11.21	12.41
Glycine (Gly)	3.82	3.70	4.18
Threonine (Thr)*	3.61	3.76	4.00
Serine (Ser)	3.00	3.10	3.45
Aspartic acid (Asp)	8.93	8.96	9.36
P-PER <sup>a</sup>	3.07	3.14	3.26

\*Essential amino acids; <sup>a</sup>P-PER= Calculated Predicted Protein Efficiency Ratio, Ra = Raw duckweed meal, Co = Cooked duckweed meal, So = Soaked in potash duckweed meal

### **Classified amino acids composition of raw and treated duckweed meals**

Treatment methods resulted in increased values of parameters for Co and So (Table 4.). Similar increasing trends were observed for total essential amino acids (TEAA) without histidine, total acidic amino acids (TAAA), and total basic amino acids (TBAA) for Ra, Co and So. However, essential aliphatic amino acids (EAAA) and essential aromatic amino acids (EARAA) varied in raw and each treatment method. Similar variations were recorded for total neutral amino acids (TNAA), total sulfur amino acids (TSAA), and percentage cysteine in TSAA (% cysteine in TSAA) for Ra, Co and So respectively.

**Table 4:** Classified amino acids composition of raw and treated duckweed meals



<b>Amino Acids (g/100g crude protein)</b>	<b>Ra</b>	<b>Co</b>	<b>So</b>
Total Amino Acids (TAA)	77.89	78.25	85.19
<b>Total Essential Amino Acids (TEAA)</b>			
With Histidine	40.03	40.24	43.67
Without Histidine	37.77	37.94	41.34
<b>% TEAA</b>			
With Histidine	51.39	51.42	51.26
Without Histidine	48.49	48.49	48.53
Total Non-Essential Amino Acids (TNEAA)	37.86	38.01	41.52
% TNEAA	48.61	48.58	48.74
Essential Aliphatic Amino Acids (EAAA)	16.99	16.60	19.08
Essential Aromatic Amino Acids (EArAA)	5.72	5.74	6.44
Total Neutral Amino Acids (TNAA)	44.09	43.94	49.27
% TNAA	56.61	56.15	57.84
Total Acidic Amino Acids (TAAA)	20.09	20.17	21.77
% TAAA	25.79	25.78	25.55
Total Basic Amino Acids (TBAA)	13.71	14.14	14.15
% TBAA	17.60	18.07	16.61
Total Sulphur Amino Acids (TSAA)	2.63	1.98	3.10
% TSAA	3.38	2.53	3.64
% Cystine in TSAA	27.00	38.38	27.42

Legend, Ra = Raw duckweed meal, Co = Cooked duckweed meal, So = Soaked in potash duckweed meal

### Essential amino acids scoring of raw and treated duckweed meals

The essential amino acids score results in Table 5 showed methionine+cysteine (TSAA) and lysine as the most limiting amino acids in raw and treated duckweed meal samples. However, leucine, valine, and threonine were found limiting in Ra and Co samples but not in So. Also, Isoleucine, Phenylalanine+Tyrosine, and tryptophan were found to be adequate in raw and treated samples. Generally, the essential amino acids composition (EAAC) in Ra and Co are lower than that of the Provisional Amino Acid (Egg) Scoring Pattern (PAAESP) (36.00), but higher in So.

**Table 5:** Essential amino acids score of raw and treated duckweed meals

EAA	PAAESP (g/100g protein)	Ra		Co		So	
		EAAC	AAS	EAAC	AAS	EAAC	AAS
Leucine	7.00	6.48	0.93	6.59	0.94	7.00	1.00
Lysine	5.50	5.04	0.92	5.35	0.97	4.80	0.87

Isoleucine	4.00	4.03	1.01	4.20	1.05	4.82	1.21
Phe+Tyr	6.00	7.83	1.31	7.62	1.27	8.77	1.46
Valine	5.00	4.56	0.91	4.59	0.92	5.01	1.00
Met+Cys (TSAA)	3.50	2.63	0.75	1.98	0.57	3.10	0.89
Threonine	4.00	3.61	0.90	3.76	0.94	4.00	1.00
Tryptophan	1.00	1.11	1.11	1.13	1.13	1.33	1.33
Total	36.00	35.29	7.83	35.22	7.79	38.83	8.76

EAA = Essential Amino Acid; PAAESP = Provisional Amino Acid (Egg) Scoring Pattern {Belschant *et al.*, (1975)}; EAAC = Essential Amino Acid Composition; AAS = Amino Acid Scores. Ra = Raw duckweed meal, Co = Cooked duckweed meal, So = Soaked in potash duckweed meal.

## Discussion

The crude protein of feed materials forms a major consideration in fish feed formulation. The crude protein of the raw and treated duckweed meal (*Lemna minor*) (Table 1) used in this study are comparable to the range of 20% to 35% reported by Appenroth *et al.*, (2017); Said *et al.*, (2022) and 28.62% to 36.25% for differently treated *L. minor* in the work of Sogbesan *et al.*, (2015). However, the results here are higher than those reported by Erdal *et al.*, (2004) 18.38% CP; and Solomon and Okomoda (2012) 17.60% CP, respectively. The observed variation in the crude protein contents in this experiment from other authors' reports could be due to the cultivation conditions of the plant as noted by Ullah *et al.*, (2022); Said *et al.*, (2022) for duckweed grown in different culture media. Xu, *et al.*, (2011) noted a decrease in the protein content with an increase in the starch content in a study where duckweed cultivated in swine medium was transferred in between fresh water and saline water. However, the 3.20% to 3.50% range of crude fat (ether extract) recorded for raw and treated duckweed (*L. minor*) meal in this experiment was lower compared to the 5.20% to 7.80% obtained in similarly treated *L. minor* in the work of Sogbesan *et al.*, (2015), 4.0% to 6.0% and 4.0% to 14.0% of the dry matter reported for various species of duckweeds by Appenroth *et al.*, (2017) and Yan *et al.*, (2013), respectively. Similarly, the range of crude fibre results of 7.70% to 8.30% obtained in the present study was higher than 4.0% obtained by Appenroth *et al.*, (2017) but lower than 35.45% reported by Solomon and Okomoda (2012), 11.0% reported by Tang *et al.*, (2015) and 13.76% to 16.15 recorded for raw and treated duckweed (*L. minor*) by Sogbesan *et al.*, (2015). Ifie *et al.*, (2020) reported that blanching increased the fiber content of DWM which was similar to cooked DWM in this study. The observed crude fiber could be attributed to the nutrients in the culture medium of the duckweed mass as noted in the works of Cui and Cheng (2015); Xu *et al.*, (2011). Zhao *et al.*, (2015) reported





an increase in the starch/crude fiber of *L. minor* in the presence of heavy metals or lack of nutrients in the culture media; while salty nutrients media have also been found to dramatically increase the fiber content of duckweed according to Sree et al., (2015). The foregoing followed Hu et al., (2022) who stated that gene expression of different DW species including culture or growth method varied their nutrient composition.

The reduction of anti-nutrient factors in plant materials has been associated with their ability to leach into the water during soaking and cooking (Soetan and Oyewole, 2009). In Table 2 the reduction of three of the six ANF was effective with the cooking method in this study. Saponins, tannins, and cyanides were significantly reduced by heat treatment of the cooking method. These results substantiate the claims of several authors on the effectiveness of heat application on many anti-nutrient substances in food ingredients. Samtiya et al., (2020) asserted that many anti-nutritional factors with toxic potential that have been measured in food are heat-stable or heat-labile. Popova and Mihaylova (2020) stated that heat application can decrease certain anti-nutrients such as phytic acid, tannins, and oxalic acid in food materials. On the other hand, soaking in potash solution as a treatment method was effective on oxalates and phytates leading to a significant reduction (70% and 77%, respectively) of their values in this study. This result is in line with Devi et al., (2018) assertion that soaking in salt solutions decreases ANF. Similarly, Agume et al., (2017) also reported that total protein, soluble sugar, and tannins in soybean flour were reduced subject to soaking as a treatment method against anti-nutrient substances. Alkaloid concentration in processed samples in Ifie et al., (2020) is similar to our study. The results of the current study, however, showed that cooking and soaking in potash were not effective on alkaloids.

Amino acids are organic compounds that combine together to form proteins and are the end products of protein digestibility in animals. The concentrated amino acid in the raw and treatment methods is glutamic acid (Table 3). The amino acid in the *Lemna spp.* Samples were within the range for *Wolffia spp.* reported by Appenroth et al., (2018). While the amino acid in DWM reported by Pagluiso et al., (2022) showed variation to our study; however, the levels ranges are comparable in values. This may be due to the geographical distribution from which the duckweeds are collected as asserted by Ifie et al., (2020). The most essential amino acid in Ra and Co is Leucine while arginine is the most concentrated in So. These results are comparable to the works of Olaofe et al., (1993), Oshodi et al., (1998), Aremu et al., (2006) as reported for legumes, and Audu and Aremu (2011) who reported similar results for differently treated red kidney bean. The values of leucine in this study compared favorably with the values reported for some legumes such

as lima bean (7.59g/100g crude protein), pigeon pea (8.40g/100g crude protein), and African yam bean (7.45g/100g crude protein) as documented by Oshodi *et al.*, (1998). Glutamic and aspartic acids (20.09g to 21.77g/100g crude protein) made up the most abundant amino acids in in the test ingredients as commonly reported in plant food samples supported by Aremu *et al.*, (2006); Adeyeye (2004); Olaofe *et al.*, (1994) and Oshodi *et al.*, (1998). The least concentrated amino acid in the raw and treated duckweed meals is cysteine (0.71g to 0.85g/100g crude protein), but with increased values in the Co and So. This was in line with the work Audu and Aremu (2011) on red kidney beans. Similarly, cooking and soaking in potash led to increasing in the concentration of some essential amino acids (EAA) such as valine (4.59 and 5.01) leucine (6.59 and 7.00), isoleucine (4.20 and 4.82) and threonine (3.76 and 4.00) over the raw values 4.56, 6.48, 4.03 and 3.61 respectively as reported for red kidney bean by Audu and Aremu (2011). Cooking and soaking also increased the concentration of serine (3.10, 3.45); glutamic acid (11.21, 12.41), and aspartic acid (8.96, 9.36) over their values for raw samples 3.00, 11.16 and 8.93 respectively. However, those treatment methods still enhanced the values of histidine (2.30 and 2.33), Arginine (6.49 and 7.02), and tryptophan (1.13 and 1.33) than raw values 2.26, 6.41, and 1.11 (EAA) with proline (2.96 and 3.01) and alanine (4.31, 4.60) above raw sample 2.81; 4.21 respectively, contrary to Audu and Aremu (2011) observation on red kidney bean. Also, cooking boosts the concentration of lysine (5.35), while lowering those of methionine (1.22), tyrosine (3.01), and glycine (3.70). The differences in the amino acids profile of raw and treated duckweed meals may be attributed to transamination and deamination reactions (Aremu *et al.*, 2009). The predicted protein efficiency ratio (P-PER), as a quality parameter used for protein evaluation (FAO/WHO, 1991), ranged from 3.07 in raw to 3.26 in soaked in potash. These values satisfy FAO requirements (FAO/WHO/UNU, 1985; Oshodi *et al.*, 1998; WHO, 2007).

The evaluation report on the classified amino acids of raw and treated duckweed meals (Table 4) showed that cooking and soaking enhanced the concentration (g/100g crude protein) in Total Amino Acids (TAA), Total Essential Amino Acids (TEAA) with histidine, without histidine and Total Non-essential Amino Acids (TNEAA) over the raw sample. The essential aromatic amino acids (EArAA) range in raw and soaked are slightly lower than the recommended range for infants (6.8-11.8) (FAO/WHO/UNU, 1985; WHO, 2007). The Total Acidic Amino Acids (TAAA) in the raw and treated samples greater than the Total Basic Amino Acids (TBAA) reveals that the protein is more acidic in nature (Aremu *et al.*, 2006). These results compare well with the work of Audu and Aremu (2011) on red kidney beans. The percentage of Total Essential Amino Acids (TEAA)



in this study ranging from 51.26% in soaked to 51.42 in cooked compares favorably with that of egg (50%) (FAO/WHO, 1991), *Vigna subterranean* concentrate (49.7%) (Aremu et al., 2008) and beach pea protein isolate (44.4%) (Chavan et al., 2011).

The most limiting essential amino acids based on their scoring (Table 5), in the raw and treated samples are lysine and methionine + cystine (TSAA) ranging from 0.87 in soaked to 0.97 in cooked and 0.57 in cooked to 0.89 in soaked respectively. These results are slightly different from Audu and Aremu (2011) report on red kidney beans, where only TSAA was found to be limiting in all their treatment methods. Leucine, valine, and threonine are also limited in raw and cooked but not in soaked. Therefore, when processing methods are considered for duckweed, there will be a need for supplementation with essential amino acids such as lysine and TSAA in soaked, but with Leucine, valine, and threonine inclusive in raw and cooked samples for healthy diets (Audu and Aremu, 2011). The crude protein and amino acids profile of raw and treated duckweed (*Lemna minor*) meals qualify it as a plant protein ingredient for fish feed formulation. However, for better protein quality, processing by cooking and soaking in potash may be inevitable.

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