

## Chemical composition of common leafy vegetables and functional properties of their leaf protein concentrates

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### Abstract

Leaves from four leafy vegetables species: *Vernonia amygdalina* (Bitter leaf), *Solanum africanum*, *Amaranthus hybridus* (Green tete) and *Telfaria occidentalis* (Fluted pumpkins) were subjected to proximate analysis and determination of energy values and nutritionally valuable minerals. Thereafter, leaf protein concentrates (LPCs) were produced from the different species using low-cost fractionation techniques. The LPCs were subsequently characterised with respect to their proximate composition, mineral constituents and functional properties. On average, the leafy vegetables contained 33.3 g/100 g DM crude protein (range 31.7–34.6 g/100 g) and 8.4 g/100 g DM (range, 7.4–9.8 g/100 g DM crude fibre. Gross energy averaged 378 k cal/100 g. The protein extracts contained, on average, 47.2 g/100 g DM crude protein (range 35.1–54.9 g/100 g) 1.4 g/100 g DM crude fibre, 7.9 g/100 g DM ether extract; 15.7 g/100 g DM ash and a gross energy of 439 kcal/100 g. Ca, Mg, Na and K were the most abundant minerals in the leaf meals and leaf protein concentrates while P and Cu were the least abundant. The fat absorption capacity (FAC) varied from 19.0±4.2% in *S. africanum* to 47.0±1.4% in *V. amygdalina* with a high coefficient of variation (CV) of 49.6%. Similarly, the water absorption capacity (WAC) varied from 149.1±4.8% in *V. amygdalina* with a CV of 55.0% while emulsion capacity and emulsion stability did not vary much, as indicated by low CVs. The foaming capacity (FC) and foaming stability averaged 8.1% (range 4.1–18.0) and 2.2% (range 2.0–2.9%), respectively. All samples had varying solubilities with change in pH. The proteins generally had multiple maxima and minima in their solubilities. The nutritive potential of the vegetables species and the dietary applications of the protein concentrates are important. © 2002 Elsevier Science Ltd. All rights reserved.

### 1. Introduction

Green vegetables have long been recognised (Byers, 1961; Oke, 1968) as the cheapest and most abundant potential sources of protein because of their ability to synthesize amino acids from a wide range of virtually unlimited and readily available primary materials such as water, CO<sub>2</sub>, and atmospheric nitrogen in sunlight. For example, cassava leaves, a by-product of cassava root harvest are, depending on the varieties, rich in protein (14–40% dry matter), minerals, vitamins B<sub>1</sub>, B<sub>2</sub>, C and carotenes (Aletor & Adeogun, 1995; Eggum, 1970; Ranvindrian & Blair, 1992).

It is well known that proteins are of prime importance to health, but they are deficient in diets of most people in the developing countries. Available literature clearly

indicate that, apart from lower methionine content, the amino acid profiles of leaf protein from most species compare favourably with those of soybean, meat, fish and egg and generally surpass the FAO essential amino acid pattern (Agbede, 2000; Barbeau, 1989; Eggum, 1970). While leaf protein extraction and utilization have been widely studied in Europe, America and Asia, the subject has remained largely under-researched and the product under-utilized on the African continent. Paradoxically, the African continent, with the highest percentage of resource-poor persons, has the most need for such cheap sources of high quality protein. This study therefore has as its main objectives, the extraction and subsequent characterisation of the leaves of common leafy vegetables and their corresponding leaf protein concentrates, with respect to their proximate composition, mineral content, gross energy and certain functional attributes.

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## 2. Materials and methods

### 2.1. Vegetables materials

All vegetables samples—*Vernonia amygdalina*; *Solanum africana*; *Amaranthus hybridus* and *Telfaria occidentalis*—were harvested from the Federal University of Technology, Akure Campus or purchased in early March of 1999 from the open market in fresh conditions. The vegetable stalks were removed and the leaves rinsed with distilled water before division into two portions. One portion was sun-dried while the other half was used for leaf protein concentrate (LPC) production.

### 2.2. Leaf protein concentrate (LPC) production

The leaves were washed and weighed prior to pulping using a Posho mill, followed by pressing using a screw press. The flow chart for the low-cost fractionation scheme, adapted from Fellows (1987) is shown in Fig. 1. The separated leaf juice was heated in batches to 80–90 °C for about 10 min to coagulate and pasteurize the leaf protein. The protein coagulum was separated from the fraction by filtering through pillow cases followed

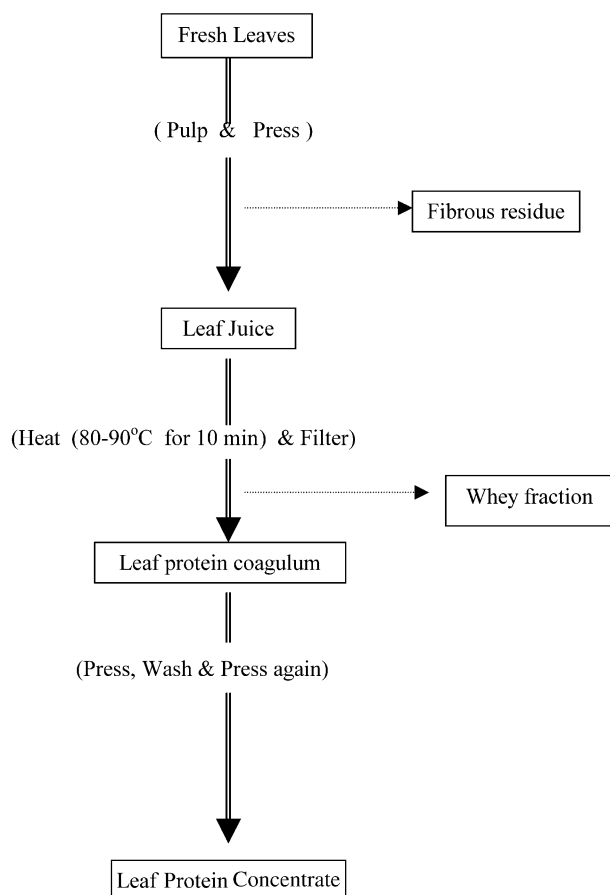


Fig. 1. Flow-chart of leaf protein concentrate (LPC) production (adapted from Fellows, 1987).

by pressing with a screw-press as described for garri making (Aletor, 1993). The LPCs were washed with water and re-pressed. The products were thereafter pulverised and spread in the sun to dry prior to use.

### 2.3. Chemical analyses

The proximate compositions of the leaves and their corresponding LPCs were determined for moisture, fat, ash and crude fibre, in triplicate, using methods described by Pearson (1976). Nitrogen was determined by the micro-Kjeldahl method, described by AOAC (1990) and the percentage nitrogen was converted to crude protein by multiplying by 6.25. The minerals were analysed after first dry-ashing at 550 °C in a Muffle furnace and dissolved in deionised water to standard volume. Sodium and potassium were determined by flame photometry while phosphorus was determined by the vanado-molybdate method (AOAC, 1990). The other minerals—Mg, Ca, Zn, Mn, Fe and Cu—were determined using an atomic absorption spectrophotometer (Vogel, 1962). The gross energy content of the different samples was computed from the proximate constituents as described by Ng and Wee (1989).

### 2.4. Determination of the functional properties of the LPCs

The protein solubilities (PS) of the leaf protein extracts were determined as described by Oshodi and Aletor (1993) and Adeyeye, Oshodi, and Ipinmoriti (1994) while the water absorption capacity (WAC) and fat emulsion stability were determined by the procedure of Beuchat (1977). The fat absorption capacity (FAC) was determined as described by Sosulski (1962). Similarly, the lowest gelation concentration (LGC), foaming capacity (FC) and foaming stability of the LPCs were determined using the technique of Coffman and Garcia (1977).

### 2.5. Data analysis

All data used were means of triplicate ( $n=3$ ) determinations. The coefficients of variation (CV) between the different families were also determined (Steel & Torrie, 1980).

## 3. Results

The mean values of the proximate composition and gross energy analysis of the leaf meal and leaf protein extracts are shown in Tables 1 and 2. The crude protein in the leaf meal (Table 1) ranged from 31.7 g/100 g DM in *V. amygdalina* to 34.6 g/100 g DM in *T. occidentalis* with a coefficient of variation (CV) of 4.5%. The mean

crude fibre content was  $8.4 \pm 1.2$  g/100 g DM with a range of 7.4 g/100 g DM in *S. africana* to 9.8 g/100 g DM in *T. occidentalis* and CV of 1.4%. Ether extract (EE) and ash averaged  $9.4 \pm 0.4$  and  $15.5 \pm 3.5$  g/100 g DM, respectively. Gross energy averaged 378 kcal/100 g with a range of 375 kcal/100 g in *S. africana* to 386 kcal/100 g in *T. occidentalis*. The leaf protein concentrates (Table 2) contained, on average,  $47.2 \pm 8.8$ ;  $7.1 \pm 3.0$ ;  $1.4 \pm 0.3$ ;  $15.7 \pm 6.2$  and  $23.8 \pm 4.2$  g/100 g DM of CP, EE, CF, ash and NFE, respectively. The crude protein content of the extract was highest in *T. occidentalis* and lowest in *A. hybridus*, with a CV of 18.6%. The major and minor nutritionally valuable mineral constituents are shown in Tables 3 and 4. Of the major minerals, Ca,

Mg, Na and K were the most abundant in the leaf meal and leaf protein concentrate. Cu and P were the least abundant minerals in all the vegetables species.

Table 5 shows the cumulative mean values (%) of fat absorption, water absorption, emulsion capacity and emulsion stability of the different leaf protein concentrates to be  $27.0 \pm 13.4$ ,  $266.5 \pm 1467$ ,  $50.0 \pm 3.2$  and  $48.0 \pm 1.2$ , respectively. Fat absorption capacity varied from  $19 \pm 4.24\%$  in *S. africana* to  $47 \pm 1.4\%$  in *V. amygdalina*, with a high CV of 49.6%. The water absorption capacity varied from  $149.1 \pm 4.8\%$  in *S. africana* to  $471.5 \pm 24.9\%$  in *V. amygdalina* with a high CV of 55.0% while EC and ES did not vary much, as shown by the low CV values of 6.5 and 2.4%, respectively.

Table 1  
Proximate compositions (g/100 g DM) of the four leafy vegetables<sup>a</sup>

Vegetable species	Family	DM	CP	EE	CF	Ash	NFE	GE
<i>S. africana</i>	<i>Solanaceae</i>	$91.8 \pm 0.3$	$34.5 \pm 1.3$	$10 \pm 0.4$	$7.4 \pm 0.2$	$17.4 \pm 1.1$	20.7	1565
<i>A. hybridus</i>	<i>Amaranthaceae</i>	$91.3 \pm 0.2$	$32.3 \pm 0.0$	$9.1 \pm 0.5$	$7.4 \pm 0.7$	$19.5 \pm 0.1$	23.0	1516
<i>T. occidentalis</i>	<i>Cucurbitaceae</i>	$91.7 \pm 0.0$	$34.6 \pm 3.2$	$9.4 \pm 0.2$	$9.8 \pm 0.9$	$13.0 \pm 0.1$	24.9	1614
<i>V. amygdalina</i>	<i>Compositaceae</i>	$91.6 \pm 0.5$	$31.7 \pm 0.0$	$9.2 \pm 0.0$	$8.8 \pm 0.2$	$12.1 \pm 0.2$	29.8	1619
	Mean	91.5	33.3	9.4	8.4	15.5	24.6	325
	S.D.	0.3	1.5	0.4	1.2	3.5	3.8	0.13
	CV (%)	0.2	4.5	4.2	1.4	2.3	15.7	27.1

<sup>a</sup> Values are for triplicate determinations. DM, dry matter; CP, crude protein; EE, ether extract; CF, crude fibre; NFE, nitrogen-free extract; GE, gross energy.

Table 2  
Proximate composition (g/100 g DM) of leaf protein extracts from the four leafy vegetables<sup>a</sup>

Vegetable species	Family	DM	CP	EE	CF	Ash	NFE	GE
<i>S. africana</i>	<i>Solanaceae</i>	$96.4 \pm 0.3$	$46.1 \pm 1.2$	$8.7 \pm 0.1$	$1.3 \pm 0.1$	$19.4 \pm 0.2$	20.3	1798
<i>A. hybridus</i>	<i>Amaranthaceae</i>	$95.5 \pm 0.2$	$35.1 \pm 1.3$	$5.6 \pm 0.3$	$1.1 \pm 0.4$	$22.3 \pm 0.1$	31.4	1584
<i>T. occidentalis</i>	<i>Cucurbitaceae</i>	$97.3 \pm 0.3$	$54.9 \pm 1.3$	$11.9 \pm 0.2$	$1.8 \pm 0.3$	$11.4 \pm 0.4$	17.3	2077
<i>V. amygdalina</i>	<i>Compositaceae</i>	$94.5 \pm 0.4$	$52.2 \pm 2.4$	$5.6 \pm 0.37$	$1.5 \pm 0.6$	$9.5 \pm 0.1$	25.7	1896
	Mean	96.0	47.2	7.1	1.4	15.7	23.7	439
	S.D.	1.2	8.8	3.0	0.3	6.2	6.6	0.3
	CV (%)	1.3	18.6	37.9	20.7	39.4	28.2	68.0

<sup>a</sup> Values are for triplicate determinations. DM, dry matter; CP, crude protein; EE, ether extract; CF, crude fibre; NFE, nitrogen-free extract; GE, gross energy.

Table 3  
Major and trace mineral components (mg/kg) of the leafy vegetables<sup>a</sup>

Vegetable species	Family	Major minerals					Trace minerals			
		Ca	Mg	Na	P	K	Fe	Zn	Cu	Mn
<i>S. africana</i>	<i>Solanaceae</i>	633	761	869	64.2	896	233	123	45.4	194
<i>A. hybridus</i>	<i>Amaranthaceae</i>	699	694	848	130	689	245	251	N.D.	415
<i>T. occidentalis</i>	<i>Cucurbitaceae</i>	521	779	975	164	838	398	254	N.D.	262
<i>V. amygdalina</i>	<i>Compositaceae</i>	524	731	896	93.4	801	127	222	39.2	144
	Mean	594	741	897	113	806	251	235	42.3	254
	S.D.	87.2	36.9	55.6	43.4	87.4	112	20.8	4.38	118
	CV (%)	14.7	4.9	6.2	38.5	10.8	44.6	8.84	10.4	46.5

<sup>a</sup> Values are for triplicate determinations.

Table 6 presents the foaming capacity and foaming stability of the leaf protein concentrates. Foaming capacity varied from  $4.1 \pm 0.0\%$  in *S. africana* to  $18 \pm 0.0\%$  in *T. occidentalis*, with high CV of 82.6%, while there was no appreciable variation in FS. Fig. 2 shows the protein solubility profiles of the leaf protein extracts. All the samples showed varying solubilities with changes in the pH. The proteins generally had multiple maximum and minimum solubilities. *V. amygdalina* had maximum solubility at pH 4. Both *A. hybridus* and *T. occidentalis* had maximum protein solubilities at pH 7 while *V. amygdalina* had maximum solubility at pH 8 and minima at pH 4 and 10.

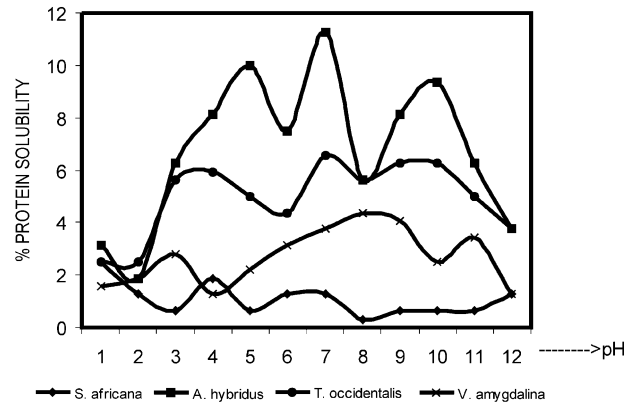


Fig. 2. Protein solubilities as a function of pH.

Table 4

Major and trace mineral components (mg/kg) of leaf protein concentrates from some common leafy vegetables<sup>a</sup>

Vegetable species	Family	Major minerals					Trace minerals			
		Ca	Mg	Na	P	K	Fe	Zn	Cu	Mn
<i>S. africana</i>	Solanaceae	688	430	518	210	658	491	366	95.3	172
<i>A. hybridus</i>	Amarathaceae	654	829	934	308	904	788	490	84.1	427
<i>T. occidentalis</i>	Cucurbitaceae	521	567	765	288	769	622	497	63.0	504
<i>V. amygdalina</i>	Compositaceae	1113	760	1005	197	974	795	422	41.3	289
	Mean	754	647	805	251	821	674	444	70.9	348
	S.D.	245	182	217	55.5	149	146	621	23.9	147
	CV (%)	32.5	28.1	26.9	22.2	18.2	21.6	13.9	33.6	42.3

<sup>a</sup> Values are for triplicate determinations.

Table 5

Absorption and emulsion capacities of different leaf protein concentrates<sup>a</sup>

Vegetable species	Family	Fat absorption (%)	Water absorption (%)	Emulsion capacity (%)	Emulsion stability (%)
<i>S. africana</i>	<i>Solanaceae</i>	19±4.2	149.1±4.8	49.1±2.2	47.8±2.3
<i>A. hybridus</i>	<i>Amarathaceae</i>	22±2.8	271.9±6.0	48.4±0.5	47.1±0.5
<i>T. occidentalis</i>	<i>Cucurbitaceae</i>	20±0	173.3±2.4	47.8±0.4	47.5±1.3
<i>V. amygdalina</i>	<i>Compositaceae</i>	47±1.41	471.5±24.9	54.8±1.4	49.7±1.4
	Mean	27	266.5	50.0	48.0
	S.D.	13.4	146.7	3.2	1.2
	CV (%)	49.6	55.0	6.5	2.4

<sup>a</sup> Values are for triplicate determinations.

Table 6

Foaming capacity and foaming stability at 30 min of different leaf protein concentrates<sup>a</sup>

Vegetables	Family	Foaming capacity	Foaming stability at 80 min
<i>S. africana</i>	<i>Solanaceae</i>	4.1±0	2.0±0
<i>A. hybridus</i>	<i>Amarathaceae</i>	4.2±0	2.0±0
<i>T. occidentalis</i>	<i>Cucurbitaceae</i>	18±0	2.0±0
<i>V. amygdalina</i>	<i>Compositaceae</i>	6.0±0	2.9±0 (at 3 h)
	Mean	8.1	2.2
	S.D.	6.7	0.5
	CV (%)	82.6	20.2

<sup>a</sup> Values are for triplicate determinations.

#### 4. Discussion

The protein values of the leaf protein extracts from these vegetables species surpass those reported by Eggum (1970) for some cassava varieties but were lower than those from leaves of some legume species reported by Oke (1968) and Agbede (2000). Subject to a high level of intake and amino acid supplementation, it is conceivable that quite a large proportion of animal protein requirement could be met by these vegetable proteins. The proximate values of these common leafy vegetables compare favourably with those of other leafy species reported by Aletor and Adeogun (1995), thus indicating their potential for use as a source of good quality food. The ash contents were fairly high, especially in *A. hybridus*. The values obtained were higher than those reported by Eggum (1970) and Oke (1968). The intake of these vegetables could be expected to contribute a large proportion of the mineral requirement in the body. The ether extract were higher than those reported for some cassava leaf species (Eggum, 1970). Deul (1955) reported that the addition of fats (ether extract) to ration lowers the specific dynamic effect of such rations and hence results in a higher energy efficiency at a given caloric intake. High nitrogen-free extract which is a measure of carbohydrate in foods, were recorded for all the samples, with *S. africana* having the least. Generally, minerals from plant sources are less bioavailable than those from animal sources (O'Del, 1969). The more important minerals involved in the building of rigid structures to support the body, i.e. calcium, phosphorus and magnesium (Osborne & Voogt, 1978) were well furnished by the vegetable species studied. These three elements in appreciable amounts, are essential for the proper formation of bones and teeth. For example, in calcium, 99% of the total amount (i.e. 1000–1200 g in adult) occurs in bones and teeth while about 600–700 g of phosphorus is also present in bones and teeth. The two elements, together with a much smaller quantity of magnesium (20–80 g), form a crystal lattice which is largely responsible for the rigidity and strength of bones and teeth.

Results on functional properties of these leaf protein concentrates clearly indicate their potential for the development of different food products. The values for water absorption capacity reported here compared favourably with the WAC reported for some seeds by Oshodi and Ekperigen (1989). Water absorption capacity values ranging from 149.1 to 471.5% are considered critical in viscous foods, such as soups and gravies. The values obtained for fat absorption capacity were rather low (19–47%) as compared with the 89.7% obtained for pigeon pea flour by Oshodi and Ekperigen (1989). This suggests that the leaf protein concentrates may not be as good as bean flour as flavour retainers. The values of

emulsion capacity (EC) and stability for the four concentrates were higher (47.8–64.8% and 47.1–48%, respectively) than the value of 7–11% reported for wheat flour (Lin, Humbert, & Sosulki, 1974). This suggests that the leaf concentrates can be used as additives for the stabilization of emulsions in the production of soups and cake. The foaming stability (Table 6) was between 2.0 and 2.9% at 30 min. These values were lower than those reported by Oshodi and Adeladun (1993) for dehulled varieties of lima bean flour whose FS ranged from 8.80 to 15.20%. The foaming stability is important since the success of whipping agents depend on their ability to maintain whip as long as possible. The four different leaf protein concentrates showed variable solubilities, with varying pH ranges, in both the acidic and basic regions, which could be useful in industrial applications. The results indicate that *A. hybridus* had greater solubility in the alkaline region which suggests its usefulness in alkaline foods. The solubility results generally indicated that leaf protein extracts from these vegetables may find good use in both acidic and alkaline foods.

Given their high proximate constituents, especially protein, and several desirable functional attributes, it is suggested that LPCs from these vegetable species may be used in enhancing the nutritional value of low-N foods such as maize gruel, yam or corn flour.

#### Uncited references

Eyo et al. (1983), Oyenuga and Tetuga (1975)

#### References

- Adeyeye, E. I., Oshodi, A. A., & Ipinmoriti, K. O. (in press). Functional properties of some varieties of African yam bean (*Sphenostylis sternocarpa*) flour. *Inter. J. Food Sci. Nutr.* 45, 829–836.
- Agbede, J. O. (2000). *Biochemical and nutritive quality of the seed and leaf protein concentrates from the seed and leaf protein concentrates from under-utilized tree and herbaceous legumes*. PhD thesis, Federal University of Technology, Akure, 2000.
- Aletor, V. A. (1993). Cyanide in garri. 1. Distribution of total, free and bound hydrocyanic acid (HCN) in commercial garri and the effect of fermentation time on its residual content. *Intern. J. Food Sci. & Nutr.*, 44, 181–287.
- Aletor, V. A., & Adeogun, O. A. (1995). Nutrient and antinutrient constituents of some tropical leafy vegetables. *Food Chemistry*, 53(4), 375–379.
- AOAC. (1990). *Official method of analysis* (14th ed.). Washington, DC, USA: Association of Official Analytical Chemists.
- Barbeau, W. E. (1989). Nutritional evaluation of experimental weaning foods prepared from green leaves, epanut oil and legume flour. *Plant Foods for Hum. Nutr.* 39, 381–392.
- Beuchat, L. R. (1977). Functional and electrophoretic characteristic of succinylated peanut flour protein. *Journal of Agricultural and Food Chemistry*, 25, 258–261.
- Byers, M. (1961). Extraction of protein from the leaves of some plants growing in Ghana. *Journal of Science and Food Agriculture*, 12, 20–30.

- Coffman, C. W., & Garcia, V. V. (1977). Functional properties and amino acid content of a protein isolate from mung bean flour. *Journal of Food Technology*, 12, 473–484.
- Deul, P. A. (1955). Sodium content of foods. *Journal of the American Dietetic Association*, 31, 21–24.
- Eggum, B. O. (1970). Protein quality of cassava leaves. *British Journal of Nutrition*, 24, 761–768.
- Eyo, S., Eyo, Mohme, & Abel, H. J. (1983). Chemical composition and amino acid contents of *Canetum africana*, *Heinsia crinita* and *Piper guineese*. *Nig. J. Nutr. Sci.*, 4:1, 55–62.
- Fellows, P. (1987). Village-scale leaf fractionation in Ghana. *Tropical Science*, 27, 77–84.
- Lin, M. J. Y., Humbert, E. S., & Sosulki, F. W. (1974). Certain functional properties of sunflower meal products. *Journal of Food Science*, 39, 368–370.
- Ng, W. K., & Wee, K. (1989). L The nutritive value of cassava leaf meal in pelleted feed for Nile Tilapia. *Aquaculture*, 83, 45–58.
- Pearson, D. (1976). *Chemical analysis of foods* (6th ed.) (pp 6–9). London: Churchill.
- O'Del, B. L. (1969). Effect of dietary components upon zinc availability: a review with original data. *American Journal of Clinical Nutrition*, 22, 1315–1322.
- Oke, O. L. (1968). Chemical changes in some Nigerian vegetables during growth. *Experimental Agriculture*, 4, 345–349.
- Osborne, D. R., & Voogt, D. (1978). *The analysis of nutrients in foods*. London: Academic Press.
- Oshodi, A. A., & Ekperingin, M. M. (1989). Functional properties of pigeon pea (*Cajanus cajan*) flour. *Food Chemistry*, 34, 187–191.
- Oshodi, A. A., & Aletor, V. A. (1993). Functional properties of hamagglutinins (lectins) extracted from three edible varieties of lima beans (*Phaseolus lunatus*). *Intern. J. Food Sci. and Nutr*, 44, 133–136.
- Oshodi, A. A., & Adedun, M. O. A. (1993). Proximate composition, some nutritional valuable minerals and functional properties of three varieties of lima bean (*Phaseolus lunatus* Linn.) flour. *Inter. J. Food Sci. Nutr.*, 43, 181–185.
- Oyenuga, V. A., & Fetuga, B. L. (1975). An overview of nutrient composition of some Nigerian vegetables. First national seminar on fruits and vegetable. In *Proc. and Recom. & papers by NIHORT*. Ibadan, Nigeria.
- Ranvindrian, V., & Blair, R. (1992). Feed resources for poultry production in Asia and the spring wheats. *Cereal Chemistry*, 39, 344–350.
- Sosulski, F. W. (1962). The centrifuge method for determining flour absorption in hard red spring wheats. *Cereal Chemistry*, 39, 344–350.
- Steel, R. G. D., & Torrie, J. A. (1980). *Principles and procedures of statistics*. Singapore: McGraw Hill, Intern. Book Coy.
- Vogel, A. T. (1962). *Quantitative inorganic analysis*. London: Longmans.