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Research Article

PHYTOCHEMICAL COMPOSITIONS AND ORGAN WEIGHT EFFECTS OF *MUCUNA SLOANEI* (*FABACEAE*) IN ALBINO RATS (*RATTUS NOVERGICUS*)

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ABSTRACT

Objectives: To assess the phytochemical composition and effects of 28 days oral administration of aqueous crude seed extracts of *Mucuna sloanei* on organ weights of normal albino rats at weekly intervals.

Materials and methods: The experiment was done using standard method. Forty eight adult male albino rats weighing between 150 to 250 g were divided into three treatment groups and one control group of twelve rats each. Each group consists of three replicates of four rats per replicate. The treatment groups were administered orally, 100 mg/kg, 200 mg/kg and 400 mg/kg of aqueous extracts of *M. sloanei* seed respectively while the control was given 1 ml/kg of normal saline using I ml syringe.

Results: The phytochemical screening of the aqueous extract showed that crude extracts of *M. sloanei* seed has flavonoids composition of (5.500±0.115), cyanide (0.054±0.001), alkaloids (1.000±0.058), tannins (0.540±0.006), phytates (14.50±0.058), saponins (6.100±0.058) and phenols (1.800±0.068) thus, having phytates as the most abundant and cyanide the least. There was no overall dose dependent and significant difference ($p>0.05$) observed in the organ weights in all the weeks when compared with the control, except the spleen, which showed a significant decrease ($P<0.05$) at 100 mg/kg at week one. Also, there was no significant difference ($p>0.05$) in the duration of treatments in the organ weights of kidney and lungs, but, a significant increase ($p<0.05$) was observed in the liver at week 4 when compared with week 1 and 2 at dose levels of 100 mg/kg. Similarly, there was an observed significant decrease ($P<0.05$) in the heart and spleen in 100 mg/kg and 400 mg/kg treated rats respectively when compared with the control.

Conclusion: The aqueous extracts of the plant seems to have antimalarial, antioxidant, antihypertensive, hypocholesterolmic and anticarcinogenic effects owing to presence of such phytochemicals as alkaloids, tannins, flavonoids, phytate, phenol and saponins. It has also shown that the plant extract did not cause inflammation or constriction at the cellular level of the organs. Thus, the *M. sloanei* seed is good for consumption.

Keywords: Phytochemical composition, Organ weight, Aqueous extract, *Mucuna sloanei*, Albino rats

INTRODUCTION

Foods of plant origin constitute the major source of food for man due to mainly their availability and low cost (Obizoba, 1998). Most people in developing countries derive their protein supplies from legumes and cereals. One of the underutilized legumes that come to mind is *Mucuna sloanei*. *M. sloanei*, commonly called "horse eye bean", is an annual leguminous climber, with pods that are covered with hairs that irritate the skin when the fruit is mature and dry (Tuleun *et al*., 2008). Its consumption by humans is localized and in many cases, it appears to be a last resort legume in circumstances of famine or scarcity of more popular legume (Ukachukwu and Obioha, 1997).

M. sloanei has been used by Igbo communities in sub-Saharan Africa as condiment or part of the main dish (Afolabi *et al*., 1985; Ukachukwu *et al*., 2002). Consumption of *Mucuna* as food has also been reported from Mozambique and Malawi (Infante *et al*., 1990; Gilbert, 2002). Seeds of *M. sloanei* are used as thickener of soup and vegetable oil in many Igbo communities of Southeastern Nigeria (Afolabi *et al*., 1985; Ukachukwu *et al*., 2002). Seeds are also used in beverages and thickening agents in recipe of several food items (Haq, 1983; Wanjekeche *et al*., 2003). The seeds are highly resistant to disease and pest and exhibit good nutritional qualities (Janardhanan and Vadiviel, 1994). Its medicinal properties include anti-diabetic (Dhawan *et al*, 1980) antiparkinsonism (Hussain and Manyam, 1997; Molloy *et al*., 2006), anti-oxidant and anti-microbial (Rajeshwar *et al*., 2005a), enhances learning and memory (Poornachandra *et al*., 2005) and antihelminthic (Jalalpure, 2007). Methanolic seed extract of *M. sloanei* has a beneficial effect on serum testosterone and LH and improved sperm count in male albino wistar rats (Egwurugwu *et al*., 2012). It may therefore be considered in the management of infertility in males.

One of the major problems with legume utilization is the presence of anti-nutritional factors (Oke *et al*., 2002). *M. sloanei* seeds have been reported to contain crude proteins, carbo-hydrates, fat, crude fibers, moisture, ash, phosphorus, magnesium, calcium, sodium, iron, manganese, copper, tannins, glycosides, L-Dopa and zinc (Giami and Wachuku, 1997; Akpata and Miachi, 2001; Ijeh *et al*., 2004; Tuleun *et al*., 2008; Nwosu, 2011). It is therefore, the aim of this study to investigate the phytochemical compositions and organ weight effects of aqueous extracts of *mucuna sloanei* (*fabaceae*) harvested in Nsukka, South Eastern Nigeria in albino rats.

MATERIALS AND METHODS

The reagents used for this research were all analytical grades

Collection and Preparation of *M. sloanei* Extract

Dried and mature nuts of *M. sloanei* were purchased from local markets around Nsukka metropolis. The seeds were identified using the identification key of Anyawu and Okoli (2004). They were dehulled, dried at room temperature and pulverized into fine powder using a milling machine. The method of extraction followed that of Akintayo *et al*. (2000). A total of 100 g of the powdered sample was introduced into 2000 ml flat bottom flask and 1500 ml of distilled water was added. The content was mixed thoroughly and left for about 24 hours with an occasional shaking to increase the extraction capacity. Thereafter, the soaked substance was filtered

with a muslin clothe (number 60 mesh size) and concentrated to dryness. The solid extract was weighed and re-dissolved in normal saline according to the body weights of the animals for oral administration.

Procurement and management of Experimental Animals

Adult male albino rats were obtained from Genetic and Animal breeding laboratory of the Department of Zoology and Environmental Biology, University of Nigeria, Nsukka. They were kept in stainless wire-rat cages equipped with drinkers and fecal collecting trays, in a clean experimental animal house. The rats were fed commercial growers chick mash (18 % crude protein) made by Vital Feeds Nigeria Limited and clean drinking water, and allowed to get acclimatized for 14 days before the start of the experiment. The animals were allowed free access to food and water *ad libitum*. The fecal droppings in the tray were removed daily. The experimental rats were handled in strict compliance with international guidelines as prescribed by the Canadian Council on the Care and Use of Laboratory Animals in Biomedical Research (1984).

Experimental Design

Forty eight (48) rats were assigned into four groups (A, B, C, & D) of 12 rats per group with each group comprising 3 replicates of 4 rats per replicate. Groups A, B, and C served as the treatment groups while group D was the control group. Three different concentrations of the aqueous extract were administered to different treatment group according to their body weights. Group A was given 100 mg/kg body weight of the seed extract while groups B and C were administered 200 mg/kg and 400 mg/kg respectively. The Control group (group D) was given 1 ml/kg body weight of normal saline. All the doses were administered once daily orally for 28 days (four weeks) to all the groups using 1 ml syringe.

Phytochemical Screening of the Crude *M. Sloanei* Seed Extracts

The quantity of some bioactive compounds such as alkaloids, cyanide, flavonoids, phenol, phytate, saponins and tannins present in the aqueous crude seed extracts of *M. sloanei* was determined using standard methods: cyanide and flavonoids was determined by the methods of Onwuka (2005); saponins and phenol by Obadoni and Ochuko (2001); alkaloids by Harbone (1973); tannins by Pearson (1976) and phyate by the methods of Oberleas (1973).

Determination of organ weights

The organs such as the heart, the liver, the kidney, the spleen, and the lungs were isolated from the anaesthetized animals and weighed using an electronic balance (Metler P C 2000) at weekly intervals for four weeks.

Statistical Analysis

The results obtained from this study were analyzed using the Statistical Package for Social Sciences (SPSS) version 17.0 for Windows. One -way ANOVA was used to test the effect of treatment and duration whereas Duncan multiple range test was used in the separation of means of the different treatment groups and duration of treatment of the same dose. All results were expressed as Mean ± Standard error of Mean (SEM), while values were considered significant at $p < 0.05$.

RESULTS

The results of the phytochemical analysis of the aqueous extract showed that *M. sloanei* seed has phytate the most abundant phytochemical and cyanide the least (Table 1). The results of the analyses carried out on the effects of the aqueous extracts of *M. sloanei* on organ weights such as Heart, Kidney, Lung, Liver and spleen at weekly intervals during the seed extract administration are shown in table 2. There was no overall dose dependent and significant difference ($p>0.05$) observed in the organ weights in all the weeks when compared with the control, except the spleen, which showed a significant decrease ($P<0.05$) at 100 mg/kg in week one. However, some minimal variations were observed at certain dose levels of the seed extracts in all the organ weights on weekly basis. Also, there was no significant difference ($p>0.05$) in the duration of treatments in the organ weights of kidney and lungs, but, a significant increase ($p<0.05$) was observed in the liver at week 4 when compared with week 1 and 2 at dose levels of 100 mg/kg (Table 2). Similarly, there was an observed significant decrease ($P<0.05$) in the heart and spleen from week 2 to 4 and week 3 to 4 in 100 mg/kg and 400 mg/kg treated rats respectively when compared with the first week (Table 2).

Table 1: Percentage Composition of Phytochemicals in the Aqueous Crude Seed Extracts of *M. sloanei*

S.No.	Phytochemicals	Composition(%)
1	Alkaloids	1.000±0.058
2	Cyanide	0.054±0.001
3	Flavonoid	5.500±0.115
4	Phenol	1.800±0.068
5	Phytate	14.50±0.058
6	Saponins	6.100±0.058
7	Tannin	0.540±0.006

Values are mean ± standard error of mean of triplicate determination

Table 2: Effects of Different Doses of the Aqueous Seed Extract of *M. sloanei* on Organ weights (g) of Albino Rats on Weekly Basis

ORGAN	CONC. (mg/kg)	DURATIONS			
		WEEK1	WEEK2	WEEK3	WEEK4
	CONTROL	0.40±0.00 ^{a,1*}	0.40±0.00 ^{a,1}	0.33±0.03 ^{a,1}	0.37±0.03 ^{a,1}
HEART	100	0.40±0.00 ^{a,2}	0.30±0.00 ^{a,1}	0.30±0.00 ^{a,1}	0.33±0.03 ^{a,1}
	200	0.43±0.09 ^{a,1}	0.43±0.09 ^{a,1}	0.33±0.03 ^{a,1}	0.37±0.03 ^{a,1}
	400	0.33±0.03 ^{a,1}	0.37±0.03 ^{a,1}	0.33±0.03 ^{a,1}	0.30±0.06 ^{a,1}
	CONTROL	0.63±0.03 ^{a,1}	0.67±0.03 ^{a,1}	0.67±0.03 ^{a,1}	0.73±0.03 ^{a,1}
KIDNEY	100	0.60±0.00 ^{a,1}	0.63±0.03 ^{a,1}	0.63±0.03 ^{a,1}	0.70±0.06 ^{a,1}
	200	0.67±0.03 ^{a,1}	0.67±0.03 ^{a,1}	0.63±0.03 ^{a,1}	0.70±0.06 ^{a,1}
	400	0.60±0.06 ^{a,1}	0.67±0.07 ^{a,1}	0.67±0.03 ^{a,1}	0.70±0.06 ^{a,1}
	CONTROL	0.60±0.00 ^{ab,1}	0.60±0.06 ^{a,1}	0.57±0.03 ^{a,1}	0.50±0.12 ^{a,1}
LUNG	100	0.60±0.00 ^{ab,1}	0.50±0.00 ^{a,1}	0.57±0.87 ^{a,1}	0.60±0.06 ^{a,1}
	200	0.63±0.03 ^{a,1}	0.63±0.00 ^{a,1}	0.60±0.06 ^{a,1}	0.73±0.15 ^{a,1}
	400	0.53±0.03 ^{a,1}	0.60±0.10 ^{a,1}	0.50±0.00 ^{a,1}	0.60±0.06 ^{a,1}
	CONTROL	3.60±0.12 ^{a,1}	3.87±0.36 ^{a,1}	3.47±0.15 ^{a,1}	3.47±0.12 ^{a,1}
LIVER	100	3.50±0.10 ^{a,1}	3.57±0.12 ^{a,1}	3.77±0.19 ^{a,1,2}	4.17±0.12 ^{a,2}
	200	3.50±0.20 ^{a,1}	3.83±0.33 ^{a,1}	3.63±0.15 ^{a,1}	4.20±0.33 ^{a,1}
	400	3.67±0.12 ^{a,1}	3.50±0.24 ^{a,1}	3.77±0.19 ^{a,1}	3.77±0.19 ^{a,1}
	CONTROL	0.40±0.00 ^{b,2}	0.37±0.03 ^{a,2}	0.27±0.03 ^{a,1}	0.27±0.03 ^{a,1}
SPLEEN	100	0.33±0.03 ^{a,1}	0.27±0.03 ^{a,1}	0.30±0.06 ^{a,1}	0.33±0.03 ^{a,1}
	200	0.40±0.00 ^{b,1}	0.30±0.00 ^{a,1}	0.33±0.03 ^{a,1}	0.30±0.10 ^{a,1}
	400	0.40±0.00 ^{b,2}	0.33±0.03 ^{a,1,2}	0.30±0.00 ^{a,1}	0.27±0.03 ^{a,1}

Values are mean ± SEM of triplicate determination. *Values with different alphabetic (lower case) superscripts differ significantly (P<0.05) between different concentrations within the same exposure duration. Similarly, values with different numeric superscripts differ significantly (P<0.05) between different exposure periods within the same concentration. Results are expressed as Mean ± SEM.

DISCUSSION

This research was done to assess the phytochemical composition of aqueous *M. sloanei* seed extracts and its possible effects on some organ weights of albino rats. The results of the phytochemical screening of the aqueous crude extract of *M. sloanei* revealed the percentage composition of alkaloids, cyanide, flavonoids, phenol, phytate, saponins and tannins with phytates being the most abundant and cyanide the least abundant (Table 1). These are the most important bioactive constituents of plant (Hill, 1952). Alkaloid is a basic natural products occurring primarily in plants. It has sedative and analgesic properties (Malu *et al*., 2009). Since *M. sloanei* contains alkaloids which possess important physiological properties, it may be used in pain relieving drugs, cough medicine, antitumor, anesthetic, analgesic etc. The extract contains insignificantly very low cyanide content (Table 1), which means that the plant seed is not poisonous, and could serve as food.

Moreover, the significant flavonoid content of the extract indicates that the plant may have antioxidant acitivity (Malu *et al*., 2009). Saponins and phenols in food medicine and masticants contribute to the phenomenon of low rate of artherosclerosis and coronary heart disease (John, 1996). Similarly, Koatkar and Rao (1997) stated that saponins in plant are recently shown to have hypocholesterolmic as well as anticarcinogenic effects. Thus, *M. sloanei* could be used in the management of cardiovascular diseases. Phytic acid has been reported to lower the nutritional value due to its limiting effects on the bioavailability of dietary minerals and essential trace elements (e.g. iron, zinc, calcium) in human intestine (Brune *et al*., 1992; Ryden and Selvendran, 1993; Gustafsson and Sandberg, 1995), they possess antioxidant, anticarcinogenic and hypoglycemic activities (Graf and Eaton, 1990; Rickard and Thompson, 1997; Shamsuddin *et al*., 1997). The significantly high level of phytate in the aqueous extracts of *M. sloanei* seed (Table 2), showed that it may have anti-cancer and anti-diabetes properties. Saponins form a group of compounds, which on consumption causes deleterious effects such as hemolysis and permeabilization of the intestine (Cheeke, 1996 and Price *et al*., 1987). This agrees with Ugwu (2012), who in their research observed that there was no significant difference in the body weight and haematological parameters of rats treated with *M. sloanei* extract. Although, tannin may decrease protein quality by decreasing digestibility and palatability, they have good antimicrobial and anti-inflammatary activities (Hertog *et al*., 1997). Therefore, the presence of this phytochemical (tannin) in the plant extract showed that the extract may play a very important therapeutic role in the field of medicine. Thus, *M. sloanei* could be used in the management of viral, bacterial and fungal diseases. The results of phytochemicals obtained in this present research corroborates the findings of Caius (1989) and Warrier *et al*. (1996) who stated that all parts of *Mucuna* are known to possess high medicinal value due to the presence of useful phytochemicals.

Organ-body weight ratio is a marker of cell constriction and inflammation (Moore and Dalley, 1999). There was an overall no significant difference in the duration of treatment in all the weights of all the organs except the heart and spleen which showed a significant decrease in the 100 mg/kg and 400 mg/kg treated rats respectively at the third and last week of administration when compared with week 1 (Table 2). Although, there was a significant increase in the liver of those animals administered 100 mg/kg at week 3 and 4 when compared with the first and second week of treatment (Table 2), we suspect that the animal may have hepatic problems which may have arisen as a result of conditions other than the *M. sloanei* extract. Similarly, except the spleen, which showed a significant reduction in week 1 at dose levels of 100 mg/kg, there was no overall significant and dose dependent effects of treatments on the organ weight ratio of the animals (Table 2). The non-significant and dose independent effect on the rat heart, kidney, lung and liver-body weight ratio following the administration of various doses of the plant extract suggests that the extract did not induce inflammation or constriction at the cellular level of such organs investigated (Moore and Dalley, 1999).

CONCLUSIONS

The aqueous extracts of the plant seems to have antimalarial, antioxidant, antihypertensive, hypocholesterolmic and anticarcinogenic effects owing to presence of such phytochemicals as alkaloids, tannins, flavonoids, phytate, phenol and saponins. It has also shown that the plant extract did not cause inflammation or constriction at the cellular level of the organs. Overall, it can be deduced from the present finding that *M. sloanei* seed is safe for consumption.

REFERENCES

1. Afolabi OA, Oshuntogun BA, Adewusi SR, Fapojuwo OO, Ayorinde FO, Grisson FE, Oke OL. Preliminary nutritional and chemical evaluation of raw seeds from *Mucuna solanei*: An underutilized food source. *Journal of Agricultural Food Chemistry* 1985; 33: 122-124.
2. Akintayo ET, Oshodi AA, Adebowale KO. Physicochemical Properties of Lima bean (*Phaseolus lunatus*) unmodified and modified starches. *Nigeria Journal of Science* 2000; 34(2) 187 – 193.
3. Akpata AO, Miachi EU. Chemical composition and selected functional properties of sweet orange and legumes flowers. *Nutrition* 2001; 54: 353-362.
4. Anyanwu PC, Okoli BE. Comparative Studies of the Morphology and Anatomy of *Mucuna Prurienes* (L.) D C and *Mucuna sloanei* (Fawc and Rendle). *Nigerian Journal of Botany* 2004; 17: 21-27.
5. Brune M, Rossander-Hulthén L, Hallberg L, Gleerup A, Sandberg AS. Human iron absorption from bread: Inhibiting effects of cereal fiber, phytate and inositol phosphates with different numbers of phosphate groups. *Journal of Nutrition* 1992; 122: 442-449.
6. Caius JF. *The Medicinal and Poisonous Legumes of India*. Scientific Publishers, Jodhpur, India; 1989.
7. Canadian Council of Animal Care. Guide to the Handling and Use of Experimental Animals.Ottawa,Canada;1984. 2. ttp://www.ccac.ca/en/CCAC_Programs/Guidelines_
8. Policies/GUIDES/ENGLISH/v2_84/CHXIX_1.HTM.
9. Cheeke PR. Biological Effects of Feed and Forage Saponins and their Impacts on Animal Production. In: Waller, G and Yamasaki, K, (Editors). Plenum Press, New York; 1996.
10. Dhawan BN, Dubey MP, Mehrotra BN, Rastogi RP, Tandon JS. Screening of Indian Plants for Biological Activity. *India Journal of Experimental Biology* 1980; 18 (9): 594-606.
11. Egwurugwu JN, Nwafor A, Olorunfemi OJ, Anaduaka SC. Effects of methanolic seed extracts of mucuna sloanei on male sex hormones and sperm quality in rats. *Journal of Medicinal Plants Research* 2012; 6(22): 3919-3922.
12. Giami SY, Wachuku OC. Composition and functional properties of unprocessed and locally processed seeds from three underutilized food sources in Nigeria. *Nutrition* 1997; 50(1): 27-36.
13. Gilbert R. *Mucuna pruriens* in Malawi: A promising legume with a troubled history. In: Flores BM, Eilittä M, Myhrman R, Carew LB, Carsky RJ. (Editors). Workshop, CIDICCO, CIEPCA and World Hunger Research Center, Tegucigalpa, Honduras; 2002.
14. Graf E, Eaton J. Antioxidant Functions of Phytic Acid Free Radical. *Biology and Medicine* 1990; 8: 61-69.
15. Gustafsson EL, Sandberg AS. Phytate reduction in brown beans (*Phaseolus vulgaris* L.). *Journal of Food Science* 1995; 60: 149-152.
16. Haq N. *New Food Legume Crops or the Tropics*. In: Nugent J, Connor MO (Editors). Cuba Foundation Symposium, 97, Pitman Books, London; 1983.
17. Harbone JB. *Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis*. Chapman and Hall, New York; 1973.

18. Hertog MGL, Sweetnam PM, Fehily AM, Elwood PC, Kromhout D. Antioxidant flavonols and ischaemic heart disease in a Welsh population of men - the caerphilly study. *American Journal of Clinical Nutrition* 1997; 65: 1489-1494.
19. Hill AF. *Economic Botany: A Handbook of Useful Plant and Plant Products*. Second Edition. McGraw-Hill Book Company Incorporation, New York; 1952.
20. Hussain G, Manyam BV. Mucuna pruriens proves more effective than L-Dopa in Parkinson's disease animal model. *Phytotherapy Research* 1997; 11: 419-423.
21. Ijeh II, Unaegbu SO, Anaga AO. Studies on some nutritional and toxicological properties of Mucuna sloanei. *African Journal of Bioresources* 2004; 2(1):24-28.
22. Infante ME, Perez AM, Simao MR, Manda F, Baquete EF, Fernandez AM. Outbreak of acute psychosis attributed to *Mucuna pruriens*. *Lancet* 1990; 336: 1129-1135
23. Jalalpure SS, Alagawadi KR, Mahajanashelti CS. In-vitro antihelmintic property of various seed oils against Pheritima posthuma. *India Journal Pharmaceutical Sciences* 2007; 69(1): 158-160.
24. Janardhanan K, Vadiviel V. Biological composition of different germplasm seed materials of India. In: Proceedings of National Seminar on Biodiversity. Strategies for conservation and future challenges. *Coimbatore India* 1994; 16-17: 93-97.
25. John T. Phytochemicals as evolutionary mediators of human nutritional physiology. *International Journal of Pharmacognosy* 1996; 34(5): 327-334.
26. Koratkar R, Rao AV. Effect of soya bean saponins on azoxymethane-induced preneoplastic lesions in the colon of mice. *Nutrition and Cancer* 1997; 27: 206-209.
27. Malu SP, Obochi GO, Edem CA, Nyong BE. Effect of methods of extraction on phytochemical constituents and antibacterial properties of *Tetracapidium conophorum* seeds. *Global Journal of Pure and Applied Sciences* 2009; 16(3/4): 373-376.
28. Molloy SA, Rowan EN, O'Brien JT, Mckeith IG, Wesnes K, Burn DJ. Effect of levodopa on cognitive function in Parkinson's disease with and without dementia and dementia with Lewy bodies. *Journal of Neurology and Neurosurgical Psychiatric* 2006; 77: 13-23.
29. Moore, K.L. and Dalley, A.F. Structure of the penis. In: Clinical Oriented Anatomy. 4th edition. Lippincot Williams and Williams; a Woller Klumner Corporation, Philadelphia; 1999. pp. 287-299.
30. Nwosu J. The effect of storage condition on the rheological/functional properties of soup thickner Mucuna sloanei (Ukpo). *Research* 2011; 3: 6
31. Obadoni BC, Ochuko PO. Phytochemical studies and comparative efficacy of the crude extracts of some home static plants in Edo and Delta States of Nigeria. *Global Journal of Pure and Applied Sciences* 2001; 8:203 -208.
32. Oberleas D. *Phytates in Toxicants Occurring Naturally in Foods*. National Academy of Sciences, Washington D.C; 1973
33. Obizoba IC. Fermented Food. In: Nutritional Quality of Plant Foods. Osagie AU, Eka, O.U. (Editors). PostHarvest Resource Unit, University of Benin City; 1998. pp. 160 -198.
34. Oke DB, Oke MO, Adeyemi OA. Predictions of cowpea seed protein quality through total sulphur determination. Proceedings of 7th Annual conference of Animal Science Association of Nig. Sept. 16-19th:121; 2002.
35. Onwuka GI. *Food Analysis and Instrumentation Theory and Practice*. Naphthali Prints, Lagos, Nigeria.; 2005.
36. Pearson DA. *The Chemical Analysis of Foods*. Seventh Edition. Churchill and living stone, Eduinburgh, London; 1976.
37. Poornachandra MN, Khanam S, Shivananda BGTN, Chivanandra TN, Dris R. Mucuna pruriens (LDDC)-A novel drug for learning and memory retrieval. *Journal of Food and Agricultural Environment* 2005; 3(3&4): 13-15.
38. Price KR, Johnson IT, Fenwick GR. The chemistry and biological significance of saponins in food and feeding stuffs. *Critical Reviews in Food Science and Nutrition* 1987; 26: 27-135.
39. Rajeshwar Y, Kumar, GPS, Gupta M, Mazumder UK. Studies on in-vitro antioxidant activities of methanol extract of *Mucuna pruriens* (Fabaceae) seeds. *Eur. Bull. Drug Research* 2005; 13: 31
40. Rickard SW, Thompson LU. *Interactions and biological effects of phytic acid*. In: Antinutrients and Phytochemicals in Food. In: Shahidi F (Editor), ACS Symposium Series # 662, American Chemical Society, Washington D.C; 1997.
41. Ryden P, Selvendran RR. *Phytic acid: Properties and determination*. In: Macrae, R, Robinson RK, Sadler MJ. (Editors). Encyclopedia of Food Science, Food Technology and Nutrition. Academic Press, London; 1993.
42. Shamsuddin AM, Vucenik I, Cole KE. A novel anticancer agent. *Life Science* 1997; 61: 343-354.
43. Tuleun CD, Carew SN, Patrick JA. Fruit characteristics and chemical composition of some varieties of velvet beans (*Mucuna spp*) found in Benue State of Nigeria. *Livestock Research and Rural Development* 2008; 20: 10.
44. Ukachukwu SN, Obioha FC. Chemical evaluation of *Mucuna conchinchinonsis* as alternative protein feedstuff. *Journal of applied chemistry and Agricultural Research* 1997; 4:33-38.
45. Ukachukwu SN, Ezeagu IE, Tarawali G, Ikeorgu JEG. Utilization of Mucuna as food and feed in West Africa. In: Flores BM, Eilitta M, Myhrman R, Carew LB, Carsky RJ. (Editors). Food and Feed from *mucuna*: Current uses and way forward. Proceedings of an International Workshop. Tegcigalpa, Honduras, April 26-29, 2002. CDICCO, CIEPCA & Judson College, Tegcigalpa, Honduras; 2002. pp. 189-217
46. Ugwu GC. Effects of aqueous extracts of *Mucuna sloanei* (fabaceae) seed on biochemical and haematological parameters of albino rats (R*attus novergicus*). *Masters degree thesis submitted in the Department of Zoology and Environmental Biology, University of Nigeria Nsukka*; 2012.
47. Wanjekeche E, Wakasa V, Mureithi JG. Effect of germination, alkaline and acid soaking and boiling on the nutritional value of mature and immature *Mucuna* (*Mucuna pruriens*) beans. *Tropical and Subtropical Agro ecosystems* 2003; 1: 183-192.
48. Warrier PK, Nambiar VPK, Ramankutty C. *Indian Medicinal Plants, a Compendium of 500 Species*. Volume 4. Orient Longman Limited, Madras, India; 1996.