INTRODUCTION

Foods of plant origin constitute the major source of food for man due to their availability and low cost (Obizoba, 1998). Most people in developing countries derive their protein supply from legumes and cereals. One of the underutilized legumes that come to mind is Mucuna sloanei. M. sloanei, commonly called “hors 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 local 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., 1997; 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 recipes of several food items (Hag, 1983; Wanjekche et al., 2003). The seeds are highly resistant to disease and pest and exhibit good nutritional qualities (Janardhanan and Vadivel, 1994). Its medicinal properties include anti-diabetic (Dhawan et al, 1980) antiparkinsonism (Hussain and Manyam, 1997; Molyko et al, 2006), anti-oxidant and anti-microbial (Rajeswar et al., 2005a), enhances learning and memory (Poornachandra et al., 2005) and antimicrobial (Jalalpore, 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 (Egyvuruygu 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 Akinbayo 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 groups 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 extract of M. sloanei was determined using standard methods: cyanide and flavonoids was determined by the methods of Ohwuka (2005); saponins and phenol by Obadoni and Ochuko (2001); alkaloids by Harbone (1973); tannins by Pearson (1976) and phytate 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.

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

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Phytochemicals</th>
<th>Composition(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alkaloids</td>
<td>1.00±0.058</td>
</tr>
<tr>
<td>2</td>
<td>Cyanide</td>
<td>0.05±0.001</td>
</tr>
<tr>
<td>3</td>
<td>Flavonoid</td>
<td>5.50±0.115</td>
</tr>
<tr>
<td>4</td>
<td>Phenol</td>
<td>1.80±0.068</td>
</tr>
<tr>
<td>5</td>
<td>Phytate</td>
<td>14.50±0.058</td>
</tr>
<tr>
<td>6</td>
<td>Saponin</td>
<td>6.10±0.058</td>
</tr>
<tr>
<td>7</td>
<td>Tannin</td>
<td>0.40±0.006</td>
</tr>
</tbody>
</table>

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

<table>
<thead>
<tr>
<th>ORGAN</th>
<th>CONC. (mg/kg)</th>
<th>DURATIONS</th>
<th>WEEK2</th>
<th>WEEK3</th>
<th>WEEK4</th>
</tr>
</thead>
<tbody>
<tr>
<td>HEART</td>
<td></td>
<td>WEEK1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CONTROL</td>
<td>0.40±0.00±1 ²</td>
<td>0.40±0.00±1</td>
<td>0.32±0.03±1</td>
<td>0.37±0.03±1</td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>0.40±0.00±1 ²</td>
<td>0.30±0.00±1</td>
<td>0.30±0.00±1</td>
<td>0.30±0.00±1</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>0.43±0.09±1 ²</td>
<td>0.30±0.00±1</td>
<td>0.33±0.03±1</td>
<td>0.37±0.03±1</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>0.33±0.03±1 ²</td>
<td>0.37±0.03±1</td>
<td>0.33±0.03±1</td>
<td>0.30±0.06±1</td>
<td></td>
</tr>
<tr>
<td>KIDNEY</td>
<td></td>
<td>CONTROL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>0.63±0.03±1 ²</td>
<td>0.67±0.03±1</td>
<td>0.67±0.03±1</td>
<td>0.73±0.03±1</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>0.67±0.03±1 ²</td>
<td>0.67±0.03±1</td>
<td>0.67±0.03±1</td>
<td>0.70±0.06±1</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>0.60±0.06±1 ²</td>
<td>0.67±0.07±1</td>
<td>0.67±0.03±1</td>
<td>0.70±0.06±1</td>
<td></td>
</tr>
<tr>
<td>LUNG</td>
<td></td>
<td>CONTROL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>0.60±0.00±1 ²</td>
<td>0.60±0.06±1</td>
<td>0.57±0.03±1</td>
<td>0.50±0.12±1</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>0.63±0.03±1 ²</td>
<td>0.60±0.00±1</td>
<td>0.50±0.06±1</td>
<td>0.73±0.15±1</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>0.53±0.03±1 ²</td>
<td>0.60±0.10±1</td>
<td>0.50±0.00±1</td>
<td>0.60±0.06±1</td>
<td></td>
</tr>
<tr>
<td>LIVER</td>
<td></td>
<td>CONTROL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>3.60±0.12±1 ²</td>
<td>3.67±0.36±1</td>
<td>3.47±0.15±1</td>
<td>3.47±0.12±1</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>3.50±0.10±1 ²</td>
<td>3.83±0.35±1</td>
<td>3.63±0.15±1</td>
<td>4.20±0.33±1</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>3.67±0.12±1 ²</td>
<td>3.50±0.24±1</td>
<td>3.77±0.19±1</td>
<td>3.77±0.19±1</td>
<td></td>
</tr>
<tr>
<td>SPILEEN</td>
<td></td>
<td>CONTROL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>0.40±0.04±1</td>
<td>0.37±0.03±1</td>
<td>0.27±0.03±1</td>
<td>0.30±0.10±1</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>0.33±0.03±1</td>
<td>0.30±0.00±1</td>
<td>0.30±0.03±1</td>
<td>0.30±0.10±1</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>0.30±0.00±1</td>
<td>0.30±0.00±1</td>
<td>0.30±0.00±1</td>
<td>0.30±0.00±1</td>
<td></td>
</tr>
</tbody>
</table>

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 as the most abundant phytochemical and cyanide as 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).
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, antiinflammatory, 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 activity (Malu et al., 2009). Saponins and phenols in food medicine and masticants contribute to the phenomenon of low rate of arteriosclerosis and coronary heart disease (John, 1996). Similarly, Kottar and Rao (1997) stated that saponins in plant are recently shown to have hypcholesterolism 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; Ricard 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., 1997). 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-inflammatory 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, hypocholesterinemic 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

8. Policies/GUIDES/ENGLISH/1284/CHXIX_LHTM.


43. Tulean 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

