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Evaluation of oxytocic and haematological effects of methanol extract of the root bark of *Spondais mombin* Linn (Anacardiaceae)

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Abstract
The present study was carried out to evaluate the oxytocic and haematological effects of the methanol extract of root bark of *Spondias mombin* Linn (Anacardiaceae) using female and male albino rats respectively. The oxytocic effect was done using isolated tissue experiment (pregnant and stilbesterol pretreated non pregnant model). The hematological study was done using whole animal experiment. The weights of the animals were determined and the blood samples collected through periorbital puncture. These were done on weekly basis for three weeks. Hematological indices of the collected blood were determined. The results of the study showed that the methanol extract of *Spondias mombin* root bark contracted the uterine horns of both the stilbesterol pretreated non-pregnant and pregnant rats just like Oxytocin and Acetylcholine. And the uterine contractility was dose dependent. Also there was a significant increase in the Red blood cells count, haemoglobin level and haematocrit level, at $p< 0.05$ of the groups that received the highest dose of the methanol extract when compared with the control. While there was a significant decrease in the white blood cell and neutrophil counts in the groups that received the extract on day 7 and the group that received the highest dose of the extract on days 14 and 21. The case is vice versa with lymphocytes. The preliminary phytochemical analysis result showed that methanol extract of root bark of *Spondias mombin* contained carbohydrates, reducing sugar, alkaloids, glycosides, Saponins, tannins, flavonoids, resins, proteins, steroids, oil and terpenoids. In conclusion, the results suggest that the root bark of this plant has oxytocic and hematological effect.

Keywords: *Spondias mombin*, phytochemical analysis, haematology, oxytocic effect

Introduction
The oxytocics are agents promoting uterine contractions. Oxytocin is the drug of choice for the induction of labour. Oxytocin is a posterior pituitary hormone that is synthesized in the hypothalamus, transported to the posterior pituitary gland for storage and then released into the circulation (Katzung, 1992). Oxytocin is a peptide secreted by the posterior pituitary gland. Oxytocin elicits milk ejection in lactating women, induces uterine contractions, maintains and augments labour. Oxytocin can be used for the control of postpartum haemorrhage. Oxytocin stimulates both the frequency and force of uterine contractions. During the first two trimesters of pregnancy, the motor activity is very low, but spontaneously the motor activity progressively increases until the sharp rise that constitutes the initiation of labour and delivery (Hardman, *et al.*, 2001) [19]. The oxytocin peptide is synthesized as an inactive precursor protein from the *OXT* gene (Sausville *et al.*, 1985, Repaske *et al*., 1990, Summar *et al*., 1990) [35, 34, 40]. This precursor protein also includes the oxytocin carrier protein neurophysin I. The inactive precursor protein is progressively hydrolyzed into smaller fragments (one of which is neurophysin I) via a series of enzymes. The last hydrolysis that releases the active oxytocin nonapeptide is catalyzed by peptidylglycine alpha-amidating monooxygenase (PAM) (Sheldrick *et al*., 1989) [36].

The use of plant and plant-based products has been a valuable and safe source omedicines for treating different kinds of diseases. This brought about the term herbal medicine, which denotes the use of herbs for their therapeutic or medicinal value (Acharya *et al*., 2008) [3]. The World Health Organization estimates that about 75% of the world populations rely on herbs to meet health care needs (WHO, 1991) [43]. Over the past decade, interest in drugs derived from plants, especially the phytotherapeutic ones, has increased expressively (Shu, 1998). It is estimated that about 25 per cent of all modern medicines are directly or indirectly derived from plants (Craig *et al*., 1997) [16]. Many plants are known to have oxytocine –like effect and they include; *Costus lucanusianus* (Owolabi *et al*., 2010) [32], *Carica papaya* (Meera *et al*., 2013), *Xyloia aethiopia* (Omodamiro *et al*., 2012) [31], *Ocimum gratissimum* (Omodamiro *et al*., 2012) [31] and *Bambusa vulgaris* (Yakubu *et al.*, 2009) [44].

Spondias mombin Linn belongs to the family Anacardiaceae. It is native to the tropical America and has been naturalized in parts of Africa, India and Indonesia and it is one of the medicinal herbs in Southern Nigeria (Aiyeloja and Bello, 2006) [6]. It is called hog plum in English (Common name), *Bal*a (Costa Rica), *Jobito* (Panama), *Jobo blanco* (Colombia), *Jobo corronchoso* (Venezuela), *Hoeboe* (Surinam) *Acaiba, Caja, Pau da tapera* (Brazil), *Ubo* (Peru), *Hobo* (Mexico) (Morton, 1987) [25]. Vernacular names; In Nigeria, it is known by various names: ogheeghe (Edo), *iyawe and Akikaetikan* (Yoruba), *akika* and ichikara (Ibo), *Tsardarmasar* (Hausa) Yoruba, *chabbuh* (Fulani) and *nsukakara* (Efik) (Uchendu, *et al*., 2008; Guill, 1992) [42]. The plant has been traditionally noted for its medicinal and food values. Preliminary results report a wide range of antibacterial and antifungal properties (Okwu, 2001) [29]. In Nigeria, the leaves are used locally by traditional medical practitioners for the treatment of different conditions such as stomach pain, cough, cuts, dizziness, eye ailments, thrush, yaw and as an expectorant. Scientific investigations have shown that it has anthelmintic, antioxidant, antimicrobial and anti-inflammatory actions (Ademola *et al.,* 2005; Abad *et al*., 1996; Abo *et al*., 1999; Calderon *et al*., 2000; Nworu *et al*., 2011; Kramer *et al*., 2002; Ademola *et al*., 2005) [4, 1, 2, 12, 26, 23]. Leaves of *Spondias mombin* have been reported to be responsible for various actions such as; smooth muscle relaxant (Akubue *et al*., 1983) [9], abortifacient (Offiah and Anyanwu, 1989) [28], sedative and anticonvulsant (Ayoka *et al*., 2006) [10], and anxiolytic (Ayoka *et al*., 2005) [11], Antiageing (Corthout *et al.,* 1992) [15], Anti-malarial (Carabolla *et al.,* 2004) [13], Anti-oxidant (Shultes and Raffant, 1990) [38], Antifertility (Raji *et al*, 2006) [33], Beta-lactamase inhibitor (Coatles *et al*, 1994) [14], Haemostatic function (Kone-Bamba *et al*, 1987) [22] and Wound healing (Ajao *et al*,, 1985) [7]. The lipid lowering effect of aqueous leaf extract of *Spondias mombin* had been demonstrated (Igwe *et al*., 2008) [20]. The root is used as source of emergency water supply although no pharmacological work with the root bark has been published so far. This study is embarked on in order to establish the oxytocic and hematological effects of the root bark as seen with the leaves.

Materials and Methods

Collection and Preparation of plants materials

The fresh root bark of *Spondais mombin* used in this experiment was collected in August at Nsukka, Enugu State, Nigeria. The root bark was identified and authenticated by Mr. Ozioko of International Centre for Ethnomedicine Drug Development (InterCEDD), Nsukka, Enugu State. The root bark was carefully separated from woody part, cut into small bits, sun-dried and pulverized using a grinder (Lab mill, Serial No. 4745, Christy and Norris Ltd., England). The powdered root bark was weighed and stored in air tight container before use.

Animals

The experimental animals used for this research study were adult Swiss albino rats (95-130 g) and mice (15-20 g) of both sexes. The animals were obtained from the animal house of the Department of Pharmacology and Toxicology, University of Nigeria Nsukka. They were housed in metal cages within the facility and allow free access to standard stock pellet and water. The animals were handled according to International Protocol for the use of Animals in experiments.

Extraction of plant material

The pulverized powdered material (500 g) was macerated with 2.5 liters of methanol and extracted at room temperature for 48 h with agitation. The filtrate was concentrated in a rotary evaporator to get the extract. The weight of extract as noted and the extract kept properly till needed.

Phytochemical analysis

The phytochemical analysis of the methanol extract was investigated using standard laboratory methods outlined by Harbone (1984) [18] and Evans (2002) [17].

Pharmacological investigation/ Evaluation

Acute toxicity test

The acute toxicity and lethality (LD_{5O}) of ME in mice (n= 13) was estimated using the method described by (Lorke, 1983) [24]. The study was carried out in two stages. In stage one, mice (n= 3) per group received oral administration of 10, 100, 1000 mg/ kg of ME and were observed for 24 h for number of deaths. At end of the 24 h, no death was recorded. Consequently a fresh batch of mice (n=1) per group received 1,600, 2,900, and 5000 mg/kg of ME respectively in the second stage of the test and were observed for 24 h for deaths and no death was recorded.

Determination of oxytocic effect of the extract

This effect was investigated using the procedure described by Akah, *et al.,* (2014) [8]. The experiment was based on the isolated tissue experiment/method using a pregnant and a stilbesterol pretreated non-pregnant Swiss albino rats. The non-pregnant Swiss albino rat was injected with Stilbesterol (0.1mg/kg) intramuscularly 24hr before the experiment. Both rats were killed. The two uterine horns of the animals were cleaned free from fatty and connective tissues and trimmed. Tubular segments of approximately equal length (2-3 cm) were removed from the uterine horns by cutting of both ends and then transferred to a petri dish containing Tyrode solution. The tissue gotten from the pregnant rat was threaded at the top and the bottom; the bottom thread was attached to the tissue- holder, while the thread is attached to the recording device. The preparation was subjected to a resting tension of 1.0 g and allowed to equilibrate for 30 – 45 minutes before it was challenged with ME at 20mg, 40mg, 80mg, 160mg and 320mg doses. Graded doses of Oxytocin (1, 2, 4, 8, 16 and 32ug) and Acetylcholine (1, 2, 4, 8, 16, 32 and 64ug) were administered;1, 2, 4, 8, 16 and 32ug to establish their effect. Subsequently, Doses of ME were added to the bath fluid sequentially, and washed out 3-4 times after the maximum responses of the tissues were attained. Conc. of bath applied methanol extract was repeated where appropriate and/or regular intervals of 3 – 20 minutes after the last washing. The ME induced responses of the uterine muscle preparations were recorded isometrically by means of the force-displacement transducer. This similar procedure was repeated for the uterine horn of stilbesterol pretreated non-pregnant rat except that different doses of oxytocin (1, 2, 4 and 8ug) and Acetylcholine (2, 4 and 8ug) were used.

Evaluation of Haematological parameters

The 20 male Swiss albino rats used for this study were obtained from the afore-mentioned location. There were grouped into four (groups A-D). They were housed in metal cages within the facility and allow free access to standard

stock pellet and water. The animals were handled according to international protocol for the use of animals in experiments. Groups A, B and C received 100, 200 and 400mg respectively while group D received water for twenty-one days (21days). Their weights were obtained before the blood collection. Blood samples were obtained from the optical plexus of the rats using a heparinized (plain) haematocrit capillary at Day 0, 7, 14 and 21 from 5:30 pm to 6:30 pm to prevent variations for analysis. The blood sample was placed in an EDTA bottle for storage and collection for experiment. The determination of the differential leucocyte count, packed cell volume, haemaglobin concentration, red blood cell count, and white blood cell count was done according to Odoh *et al.* (2016) [27].

Differential Leucocyte Count

A thick blood film was made on a grease free microscope slide and was allowed to dry. The dry blood film was stained with leishman stain and was washed off after ten minutes and was allowed to air-dry. The prepared slide was viewed in the microscope, while the neutrophils and the lymphocytes were counted and their percentage composition calculated.

Packed cell volume

Haematocrit capillary tube was filled with blood by placing one of the open ends of the tube in the blood bottle and tilting it at an angle about 30º. One end of the filled capillary tube was sealed with a plastercine and the tube centrifuged for 20minutes at 300 rpm in a haematocrit centrifuge. A haematocrit reader was used to read off the length of the packed cell in percentage

Haemaglobin concentration

Sahli haemoglobinometre was used for the determination of haemoglobin concentration. Up to the ten (10) mark of the sahli tube was 0.1N Hcl placed. With the Sahli blood pipette, 20µl of blood was placed into the Sahli tube and was sucked up and down. The mixture was allowed to stand for 5minutes for the formation of acid haematin. The dark mixture formed was diluted gradually with distilled water till the colour when compared with that in the haemoglobinometre is slightly darker than the standard. The dilution continued till the colour turns exactly and slightly paler than the standard. The volumes of the noted colour change were taken and the average of the of the slightly darker and paler were compared and was ensure that the variation is not more than ±5 for the average value to be adopted. The average value was applied in the formular below:

X (Y)/100 where X is the 14g Hb in 100ml of blood, y is the average value of dilutions.

Red blood cell count

The experiment was done using the procedure described by (David, 2009). Using the dilution pipette with RED mixer from hemacytometer kit, blood was drawn up to the 0.5 mark. With the pipette left in horizontal position as possible, Ringer's solution diluents was drawn up to the 101 mark. (Dilution of 1 to 200). The tip of the pipette was seal with the finger and shaked well to mix. Half of the content of the pipette was emptied into a waste container and a small amount of the diluted blood was placed into one chamber of the hemacytometer to just fill the chamber of the hemacytometer. The preparation was allowed to sit for a minute (for cells to settle). The center of the grid was focused with 100x objective, and was counted with 400x objective. The count of each five fields (each with 16 smallest squares) with a clicker (fields: top Right & Left, bottom Right & Left, center) were noted. And all the cells touching left and bottom sides were counted ignoring the cells touching top and right sides. The RBCs/mm was calculated by adding the cells in the 5 groups and multiplying by 10,000 (i.e., add four zeros).

White blood cell count

Same as in RBC excerpt that the diluting fluid is 1.5% acetic acid tinted with methyl violet. The pipette is similar but with different graduation. Unlike the RBC, the leucocytes cells in the entire 9 big grid was counted and applied in the formular n X 200/9

Statistical Analysis

Statistical analyses of the hematological data were performed using SPSS, Data were presented as means (± SEM). Statistical differences were determined using one-way analysis of variance (ANOVA; 95% Confidence Interval), followed by Dunnett's post hoc test (Dunnett and Goldsmith, 1993). In all cases, the statistical significance was established at values of $P< 0.05$.

Results

Percentage yield of the extract: The extraction process yielded 20% of *Spondias mombin* methanol root bark extract.

Phytochemical constituents of extract

The phytochemical analysis showed that the root bark of *Spondias mombin* plant contains carbohydrates, saponins, alkaloids, tannins, reducing sugars, glycosides, flavonoids, terpenoids, proteins, oils, steroids, and resins (Table 1).

Table 1: The result of phytochemical screening of the methanol extract of *Spondias mombin* root bark.

Constituent	Inference
Carbohydrates	+
Saponins	+
Alkaloids	+
Tannins	+
Reducing sugars	+
Glycosides	+
Flavonoids	+
Terpenoids	+
Proteins	+
Oils	+
Steroids	+
Resins	+

Key: - = Absent, + = Present

Acute toxicity test

The result showed that the extract showed no death in the first 24 h among the three groups of mice that received 10, 100 and 1000 mg/kg (Table 2). Also, no death occurred at the end of the next 24 hour with second set of mice that received 1600, 2900 and 5000 mg/kg. Hence, the extract is safe and has a wide range of effective dose.

Table 2: Acute toxicity (LD_{50}) of the methanol extract of *Spondias mombin* root bark

Stage	Extract	Dose (mg/kg)	Mortality	Observation
1	Methanol	10	0/3	Find below
		100	0/3	-
		1000	0/3	-
2	Methanol	1600	0/1	-
		2900	0/1	-
		5000	0/1	-
	Control	-	0/1	-

The group of mice that received 10 mg/kg expressed normal affect during the period of the experiment. While those given 100 and 1000 mg/kg were a bit restless within the first few minutes and later became calm throughout the experiment. And the mice that received 1600 and 2900 mg/kg respectively experienced depression for 20 min and later adjusted, maintaining normal affect throughout the experiment. The mouse that received 5000 mg/kg experienced itching in the mouth and was restless for 30min, though they subsided after 2 h.

Oxytocic effect of the extract

Oxytocin, Acetylcholine, and methanol extract of *Spondias mombin* root bark induced dose related increase in force of contraction of the rat-isolated uterus.

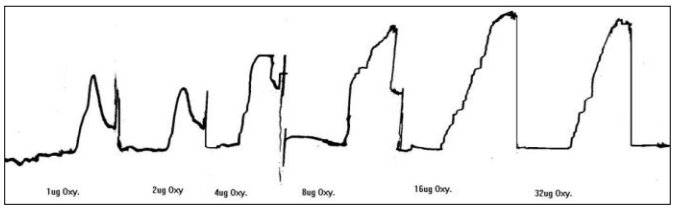

Fig 1: Effect of Oxytocin on rat uterus (pregnant)

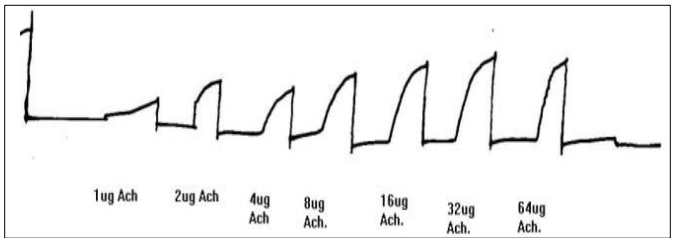

Fig 2: Effect of Acetylcholine on the rat uterus (pregnant)

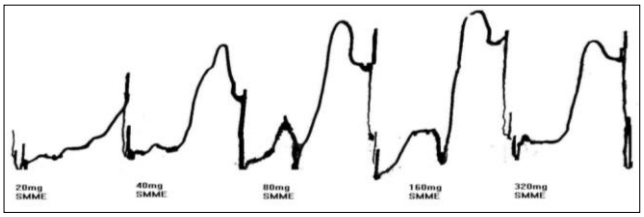

Fig 3: Effect of methanol extract of root bark of *Spondias mombin* on rat uterus (pregnant)

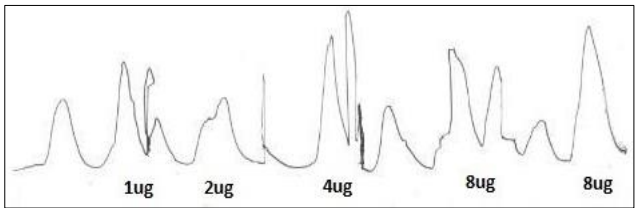

Fig 4: Effect of Oxytocin on a rat uterus (non-pregnant)

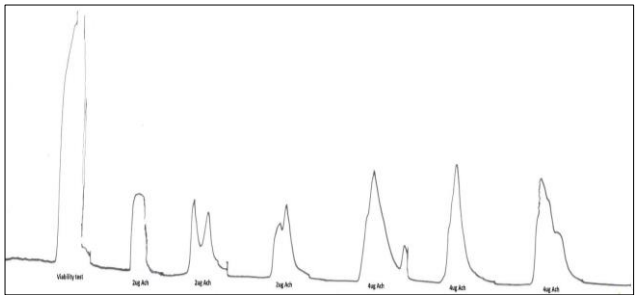

Fig 5: Effect of Acetylcholine on a rat uterus (non-pregnant)

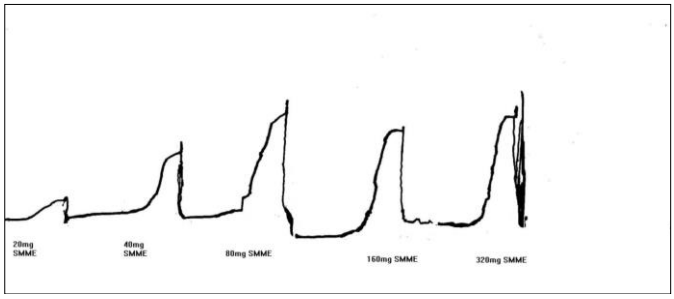

Fig 6: Effect of methanol extract of root bark of *Spondias mombin* on a rat uterus (non-pregnant)

Haematological studies

Hematocrit

The group of rats that received the highest dose of methanol extract i.e. group C had a significant increase compared to the control after first and second week of extracts administration (Table 3). There were no significant differences in treatment group A, B when compared with the control.

Table 3: Effect of methanol extract of *Spondias mombin* on hematocrit (%)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100mg/kg)	29.80 ±0.37	31.60±0.93	32.50±1.19	38.50±1.32
B (200mg/kg)	30.20±0.66	32.00±1.34	33.40±1.29	42.20±2.13
C (400mg/kg)	32.20±1.28	37.40±1.69 *	37.00±1.82 *	44.60±1.89
D (vehicle)	31.20±1.74	30.60±1.29	30.80±1.53	38.80±1.16

Values represented as Mean ± SEM of the results from triplicate analysis, $p<0.05$ significant, comparison with control group

RBC count

It can be seen that at Days 7, 14 and 21, group C had a signficant increase when compared to the control group (Table 4).

Table 4: Effect of methanol extract of *Spondias mombin* on RBC count (10^6cells/mm^3)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100 mg/kg)	3.17±0.08	3.09±0.17	3.14±0.18	3.40±0.28
B (200 mg/kg)	3.45±0.10	3.16±0.07	3.27±0.22	4.13±0.33
C (400 mg/kg)	3.31±0.17	3.72±0.1 *	3.05±0.23 *	4.56±0.22 *
D (vehicle)	3.23±0.12	2.89±0.65	2.82±0.12	3.23±0.33

Values represented as Mean ± SEM of the results from triplicate analysis, $p< 0.05$ significant, comparison with control group

Hemoglobin

The result shows that the group received highest dose of methanol extract on days 7, 14 and 21, had a significant increase when compared with the control group (Table 5a).

Table 5a: Effect of methanol extract of *Spondias mombin* on heamoglobin (g/dl)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100mg/kg)	10.58±0.24	11.48±0.38	11.08±0.44	13.50±0.46
B (200mg/kg)	10.64±0.28	11.08±0.41	11.28±0.35	14.28±0.54
C (400mg/kg)	11.06±0.42	12.96±0.60 *	12.56±0.57 *	14.70±0.37 *
D (vehicle)	10.80±0.60	10.54±0.36	10.52±0.49	12.86±0.47

Values represented as Mean ± SEM of the results from triplicate analysis, $p<0.05$ significant, comparison with control group

WBC cell count

The result shows that there was a significant difference in all groups that received the extract on Day 7 and the group that received the highest dose of the extract on days 14 and 21 against the control (Table 5b).

Table 5b: Effect of methanol extract of *Spondias mombin* on WBC count (10^3cells/mm^3)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100 mg/kg)	6.40±0.23	7.04±0.2 #	6.88±0.41	5.99±0.47
B (200 mg/kg)	6.44±0.34	6.31±0.18 #	6.51±0.24	5.67±0.30
C (400 mg/kg)	6.50±0.32	5.77±0.45 #	6.02±0.27 #	5.07±0.41 #
D (Vehicle)	6.64±0.29	8.33±0.30	7.10±0.12	6.80±0.15

Values represented as Mean ± SEM of the results from triplicate analysis, $p< 0.05$ significant, comparison with control group.

Neutrophils

The result shows that there was a significant difference with the groups that received various doses on the 7th day and the group that received highest dose of the extract on days 14 and 21 against the control (Table 6).

Tables 6: Effect of methanol extract of *Spondias mombin* on Neutrophil count (%)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100 mg/kg)	70.20±1.69	73.60±1.47 #	74.00±1.41	71.25±1.25
B (200 mg/kg)	71.00±0.84	70.60±1.78 #	72.20±1.02	64.20±3.17
C (400 mg/kg)	71.00±1.10	69.80±1.20 #	69.40±1.60 #	57.40±1.66 #
D (Vehicle)	72.00±0.71	76.80±1.36	76.40±1.33	71.20±2.73

Values represented as Mean ± SEM of the results from triplicate analysis, $p< 0.05$ significant, comparison with control group

Lymphocyte count

The result showed that there was significant increase of all the groups on the 7th day and the group that received highest dose of the extract on days 14 and 21 against the control (Table 7).

Table 7: Effect of methanol extract of *Spondias mombin* on lymhocyte count (%)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100 mg/kg)	29.80±1.69	26.40±1.47 #	26.00±1.41	28.75±1.25
B (200 mg/kg)	29.00±0.83	27.40±1.25 #	27.80±1.02	35.80±3.17
C (400 mg/kg)	29.00±1.10	30.20±1.20 #	30.60±1.60 #	42.60±1.66 #
D (Vehicle)	28.00±0.71	23.20±1.36	23.60±1.33	28.80±2.73

Values represented as Mean ± SEM of the results from triplicate analysis, $p< 0.05$ significant, comparison with control group

Effect of the methanol extract on the weight of the rats

The result shows that there were no significant difference between weights of the groups that received the methanol extract and the control (Table 8).

Table 8: Effect of methanol extract of *Spondias mombin* on the weight of the rats (g)

Treatment group	Day 0	Day 7	Day 14	Day 21
A (100 mg/kg)	104.14±5.95	118.78±5.67	133.96±6.78	147.10±5.88
B (200 mg/kg)	104.88±5.67	115.31±1.42	134.44±5.47	140.07±4.52
C (400mg/kg)	113.20±7.40	121.45±8.18	137.58±8.55	134.46±7.54
D (Vehicle)	104.80±5.38	114.22±3.61	129.20±5.62	126.57±3.85

Values represented as Mean ± SEM of the results from triplicate analysis, $p< 0.05$ significant, comparison with control group,

Discussion

After the extraction process the percentage yield was 20% and the phytochemical analysis indicated the presence of saponins, carbohydrates, alkaloids, tannins, flavonoids, glycosides, terpenoids, steroids, oils, proteins, resins and reducing sugars. "Saponins has relationship with sex hormone involved in controlling the onset of labor in women and the subsequent release of milk" (Okwu and Okwu, 2004) [30]. With this fact we can attribute its oxytocic effect to the presence of saponins in the methanol root bark extract.

This study has established the oxytocic effect of the methanol extract of *spondias mombin* root bark because the extract was able to contract the uterine horn of both the pregnant and stilbesterol pretreated non-pregnant swiss albino rats as was seen with Oxytocin and Acetylcholine. The fact that the contractile effect is dose related showed the potency of the extract in contracting the uterus. The Oxytocin and the Acetylcholine exhibited high potency than the methanol extract. This may be attributed the purity of the drug while the extract in its crude and un-purified state contains different constituents, some of which may even have antagonistic effect.

The rat uterus tissue was used in ethnopharmacological screening because *Spondias mombin* herbal remedy is used in stimulating childbirth traditionally. The smooth muscle of the rat uterus was the point of contact for these herbal drugs experimentation due to its high sensitivity among other laboratory animals. The methanol extract of *Spondias mombin* contracted and increased the uterine motility.

The use of medicinal plants to facilitate labour may be due to stimulation of muscarinic receptors in the uterine tissue or through the synthesis and release by prostaglandins well known to be myometrial stimulants reported to mediate the activity of most drugs that stimulate uterine contraction (Solloff, 1979) [30].

In the interpretation of the result, normal motility is the baseline for that particular tissue. Standard drugs were used as the controls of the set up. The medicinal plant selected for ethnopharmacological tests were based on the ethnobotanical indigenous knowledge. The fact that traditional healers have been using this plant is a worthwhile reason to investigate their efficacy in the claimed use and matching preparations in the laboratory with indigenous knowledge. The study also established the hematinic effect of the extract. There was a significant increase in the hematocrit, Red Blood Cell and hemoglobin count in the group of rats that received the highest dose of the extract when compared with the control at $p< 0.05$. The animals gained weight, remained healthy and no death was recorded throughout the experimental period and no significant difference with the control. The increase in body weight of all groups weekly were considered normal and gradually as observed in rats of similar age group in a published reference (Taconomic Technical Library, 2011) [41]. The increases in body weight were in line with the increase in food and water consumed by the rats.

Defective haematopoiesis is indicated by reduction in erythrocytes number and hemoglobin content. However, in this study, there was no indication of anemia from the levels of hemoglobin found. Hemoglobin is a protein used by red blood cells to distribute oxygen to other tissues and cells in the body. An increase in RBC count may be attributed to the direct stimulation on hemopoietic tissues such as the liver and bone marrow by the extract. Since haematocrit is a measure of the volume of RBC over the total blood volume, significant increase in haematocrit is expected since there was significant increase in the RBC count. And this increase may be indicative of normal functioning of the bone marrow in the process of erythropoesis.

There was a significant reduction in the white blood cell and neutrophil count. The hematological effect of the extract is dose related as the significant differences were seen with the group that received the highest dose of the extract except on the 7th day where all the groups had a significant decrease

when compared with the control. This effect was a vice versa for the lymphocyte count.

Conclusion

In conclusion, the methanol extract of *Spondias mombin* root bark produced oxytocic effect in both (pregnant and stilbesterol pretreated non-pregnant Swiss albino rats) and also haematinic effect. The oxytocic effect may be attributed the saponins and alkaloid present in the plant part. While the phytoconstituent(s) responsible for the hematinic effect is/are unknown. Hence further studies are needed to identify, isolate and characterize the phytoconstituent(s) of the plant part responsible for that effect. From the foregoing, the root bark extract can be very beneficial in obstetrics when constituents are isolated and further purified and also in treating anemic conditions.

References

1. Abad MJ, Mermejo P, Carretero E, Martinez-Acitores C. Anti-inflammatory activity of some medicinal plant extracts from Venezuela. Journal of Ethnopharmacology. 1996; 55(1):63-68. Doi: 10.1016/s0378-8741(96)01478-X
2. Abo KA, Ogunleye VO, Asindi JS. Antimicrobial potential of *Spondias mombin*, Croton zambesicus and Zygotritonia crocea. Phytothe Res. 1999; 13:494-497. PMID: 10479760
3. Acharya NA, Deepak M, Shrivastava A. Indigenous Herbal Medicines. Tribal Formulations and Traditional Herbal Practices, Aavishkar Publishers Distributor, India, 2008, 440-520
4. Ademola IO, Fagbemi BO, Idowu SO. Antihelmintic activity of extract of *Spondias mombin* against gastrointestinal nematodes of sheep: Studies *in vitro* and *in vivo*. Trop Anim, Health Prod. 2005; 37(3):223-235. DOI: 10-1023/ B: TROP. 0000049296.47350.80
5. Aiyeloja AA, Opeyemi IA. Non-timber forest products' marketing in Nigeria. A case study of Osun state (PDF online reproduction). Educational Research and Reviews s.l.: Academic Journals. 2006; 1(2):52–58. ISSN 1990-3839. OCLC 173185259.
6. Aiyeloja AA, Bello OA. Ethnobotanical potentials of common herbs in Nigeria: A case study of Enugu state. Ed Res Rev. 2006; 1(1):16-22.
7. Ajao AO, Shonukan O. Antibacterial effect of aqueous and alcohol extracts of *Spondias mombin* and *Alchomea cordifolia*: 2 local antimicrobial remedies. International Journal of Crude Drug Research. 1985; 23:67-72.
8. Akah PA, Ezike AC, Okoye TC, Nworu CS, Nwabunike SI, Mbaoji NF *et al*. Experimental methods in physiology (for Medical and pharmacy Students) Book III, 2014, 40-42.
9. Akubue PI, Mittal GC. Aguwa CN. Preliminary Pharmacological study of some Nigerian medicinal plants. Journal of Ethnopharmacology. 1983; 8:53-63. PMID: 6632737
10. Ayoka AO, Akomolafe RO, Iwalewa EO, Akanmu MA, Ukponmwan OE. Sedative, antiepileptic and antipsychotic effects of *spondias mombin* L. (Anacardiacaea) in mice and rats. Journal of Ethnopharmacology. 2006; 103:166-175.
11. Ayoka AO, Akomolafe R.O, Iwalewa EO, Ukponmwan OE. Studies on the anxiolytic effect of *Spondias mombin* L. (Anacardiaceae) extracts. J trd. CAM. 2005; 2(2):153-165. ISSN: 0189-6016
12. Calderon AI, Angerhofer CK, Pezzuto JM, Famsworth NR, Foster R. Forest plots as a tool to demonstrate the Pharmaceutical Potential of Plants in a Tropical Forest of Panama. Econ. Bot. 2000; 53(3):278-294.
13. Caraballo A, Caraballo B, Rodriguez-Acosta A. Preliminary assessment of medicinal plants used as antimalarials in the South-Eastern Venezuelan used as antimalarials in the South-Eastern Venezuelan Medicina-Tropical. 2004; 37(2):186-188.
14. Coates NJ, Gilpin ML, Gwynn MN, Lewis DE, Milner PH, Spear SR *et al*. SB-202742 a novel beta-lactamase inhibitor isolated from *Spondias mombin*. Journal of Natural Products. 1994; 57:654-657.
15. Corthout J, Pieters LA, Claeys M, Vanden Berghe DA, Viletinck AJ. Antiviral caffeoyl; Esters from *spondias mombin.* Phytochemistry. 1992; 31:79.
16. Craig GM, Newman DJ, Snader KM. Natural products in drug discovery and development. J Nat Prod. 1997; 60:52-60. PMID: 9014353
17. Evans WC. Trease and Evan pharmacognosy. 14th edition, London: W. B. Saunders, 2002, 33.
18. Harborne JB. The flavonoids. In, Phytochemical Methods. London: Chaman and Hall Ltd., 1984, 49-188.
19. Hardman JG, Limbird LE, Goodman-Gilman A. Goodman and Gilman The Pharmacological Basis of Therapeutics. 10th Edition. MacGraw-Hill, Medical ublishing Division, 2001.
20. Igwe CU, Ojiako OA, Nwaogu LA, Onyeze GOC. Lipid lowering effect of aqueous leaf extract of *Spondias mombin*. Linn. The internet J Pharmacol. 2008, 6(1). ISSN:1531-2976
21. Katzung BG. Histamine, Serotonin and the Ergot Alkaloids. In Katzung B.G., Masters S.B., and Trevor A. J. In Basic and Clinical Pharmacology 12th edN., 2012.
22. Kone-bamba D, Pelissier Y, Ozoukou ZF, Kouao D. A study of the haemostatic activity of fifteen medicinal plants of the traditional pharmacopoeia of Ivory Coast. Plantes-Medicinales-et-Phytotherapie. 1987; 21(2):122-130.
23. Kramer A, Mosquera E Ruiz J, Rodriguez E. Ethnobotany and biological activity of plants utilized during pregnancy and child birth in the Peruvian amazon. Emanations from the rainforest of Carribean, Cornell University, 2002, 4.
24. Lorke D. A new approach to practical acute toxicity testing. Arch. Toxicol. 1983; 54(4):257-287. PMID: 6667118
25. Morton J. Yellow mombin. In Fruits of warm climates. Julia F. Morton, Miami, Florida, 1987, 245-248.
26. Nworu CS, Akah PA, Okoye FB, Toukam DK, Udeh J, Esimone CO. The leaf extract of *Spondias mombin* L. displays an anti-inflammatory effect and suppresses inducible formation of tumor necrosis factor-α and nitric oxide (NO). J Immunotoxicol. 2011; 8(1):10-6.
27. Odoh UE, Onugha VO, Chukwube VO. Evaluation of antidiabetic effect and hematotological profile of methanol extract of *Ceiba pentandra* G (Malvaceae) stem bark on alloxan-induced diabetic rats. African Journal of Pharmacy and Pharmacology. 2016; 10(28):584-590.
28. Offiah VN, Anyanwu II. Abortifacient activity of an aqueous extract of *Spondias mombin* leaves. J Ethnopharm. 1989; 26:317-320. PMID: 2615412
29. Okwu DE. Evaluation of Chemical Composition of indigenous species and flavouring agents. Global. J Pure Appl. Sci, 2001, 455-460. ISSN: 1118-0579

30. Okwu DE, Okwu ME. Chemical Composition of *Spondia mombin* plants J Sustain. Agri. Environ. 2004; 6:140-147.
31. Omodamiro OD, Oheri OC, Nweke IN. Oxytocic effect of aqueous,ethanolic, n-hexane, and chloroform extracts of *Xylopia aethiopia*,(Annonaceae) and *Ocimum gratissium*,(Labiate) on guinea pig uterus, 2012, 40.
32. Owolabi OJ, Omogbai EK, Falodun A. Oxytocic effects of the aqueous leaf extract of *costus lucanusianus* on isolated non-pregnant rat uterus. Pak J Pharm Sci. 2010; 23(2):207-11.
33. Raji Y, Gbadegesin MA, Osonuga OA, Adisa RA, Akinsomisonye OS, Awobajo FO, Kunle-Alabi OT *et al*. Reproductive, HHaematologic and Biochemical profile of male rats treated with aqueous extract of *Spondias mombin* bark, International journal of Pharmacology. 2006; 2(1):126- 130.
34. Repaske DR, Phillips JA, Kirby LT, Tze WJ, D'Ercole AJ, Battey J. Molecular analysis of autosomal dominant neurohypophyseal diabetes insipidus. The Journal of Clinical Endocrinology and Metabolism. 1990; 70(3):752-7. doi:10.1210/jcem-70-3-752. PMID 1968469.
35. Sausville E, Carney D, Battey J. The human vasopressin gene is linked to the oxytocin gene and is selectively expressed in a cultured lung cancer cell line. The Journal of Biological Chemistry. 1985, 260(18):10236–41. PMID 2991279.
36. Sheldrick EL, Flint AP. Post-translational processing of oxytocin-neurophysin prohormone in the ovine corpus luteum: activity of peptidyl glycine alpha-amidating mono-oxygenase and concentrations of its cofactor, ascorbic acid". The Journal of Endocrinology. 1989; 122(1):313-22. doi:10.1677/joe.0.1220313. PMID 2769155.
37. Shu YZ. Recent natural products based drug development. A pharmaceutical industry perspective. J Nat prod. 1998; 61:1053-107. PMID: 9722499.
38. Shultes RE, Raffauf RF. The healing forest: Medical and toxic plants of the Northwest Amazonia, Dioscorides Press, Portland, Oregon, 1990.
39. Sollof MS. Regulation of oxytocin action at the receptor level. Life Science. 1979; 25:1453.
40. Summar ML, Phillips JA, Battey J, Castiglione CM, Kidd KK, Maness KJ *et al*. Linkage relationships of human arginine vasopressin-neurophysin-II and oxytocin-neurophysin-I to prodynorphin and other loci on chromosome 20. Molecular Endocrinology. 1990; 4(6):947–50. doi:10.1210/mend-4-6-947. PMID 1978246.
41. Taconomic Technical Library for Plants: *Carica papaya* was originally described and published in *Species Plantarum* 2:1036. 1753. GRIN *Carica papaya* information from NPGS/GRIN. National Germplasm Resources Laboratory, Beltsville, Maryland: USDA, ARS, National Genetic Resources Program. Retrieved, 2011-2015.
42. Uchendu CN, Isek T. Antifertility activity of aqueous ethanol leaf extract of *Spondias mombin* (Anacardiaceae) in rats. Afr. Health Sci. 2008; 8(3):163-167.
43. World Health Organization. Traditional medicine and modern health care. Progress report by the Director General, Document A. WHO, Geneva. 1991; 44(10):22.
44. Yakubu MT, Bukoye BB. Abortifacient potentials of the aqueous extract of *Bambusa vulgaris* leaves in pregnant Dutch rabbits, 2009.