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Anti-Inflammatory and Sedative Effects of Intra-Peritoneal Administration of Methanolic Extract of *Securidacalonge pedunculata* Root in Albino Rat

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Abstract

Objectives: The methanolic extract of the root bark of *Securidaca longipedunculata* Fres (*Polygalaceae*) was evaluated for anti-inflammatory and sedative activities in albino wistar rats using intraperitoneal route.

Methodology: Anti-inflammatory activity was determined by inducing inflammation in the paw of rats by sub-plantar injection with fresh undiluted egg albumen and after thirty minutes, inhibition of inflammation was assessed by changes in paw diameter at hourly intervals. Sedative activity was assessed by determining the effect of the extract on length of diazepam-induced sleeping time.

Results: Results of the present study revealed that, the extract possess anti-inflammatory and sedative activities in the third hour.

Conclusion: The extract has been established to have analgesic effect, and in this study to have anti-inflammatory property with central sedative effect. This indicates that the extract may have analogous mechanism of action with narcotic analgesics. The study therefore affirms the traditional use of the plant for the same purpose and can be adopted as an alternative therapy in various inflammatory conditions.

Keywords: *Securidaca longipedunculata*; Methanol extract; Anti-inflammatory; Sedation; Hypnosis; Analgesics; Diazepam

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Introduction

Research exploration for newer pharmacologically active compounds from plants of indigenous African regions is on the increase [1]. Utility of plants in ethno-medicine is often used as guide for screening its activity to confirm the claim. Compounds with reputed activity are subjected to further scrutiny that could eventually lead to drug development. In addition, knowledge of the possible mode of action could give a scientific rationale for integration of complementary medicine and orthodox medicine. *Securidaca longipedunculata* Fres (*Polygalaceae*) is shrub with twisted bole or slender erect branches and grows up to 30 feet high [2], that grows in savannah vegetation. It is used among Hausa People of Nigeria as an analgesic and also in treating many

disease conditions [3]. As such, its vernacular name on translation implies mother of medicines [4]. Oral administration of the aqueous root extract of plant has been scientifically investigated to have analgesic activity [5]. Methanolic extract of both the root and stem of the plant also exhibited similar analgesic properties [6]. Bioavailability of some pharmacologically active compounds is affected by absorption loss and first pass effect using oral route [7]. It is therefore unknown if active ingredient of *Securidaca longipedunculata* responsible for analgesic activity is degraded by oral route. Screening the extract for other collateral activity of known analgesics would give an insight into the nature of the compound(s) responsible for the observed analgesic activity. This study aimed at determining the anti-inflammatory and sedative effect of crude methanol extract of *Securidaca*

longipendunculata in albino rats using a parenteral route of administration.

Materials and Methods

Plant collection and identification

Securidaca longipendunculata root bark was obtained in the bush near Ngulde in Askira/Uba Local Government Area of Borno State. It was identified and authenticated in the Department of Biological Sciences, University of Maiduguri, and a voucher sample (Vet212A2) was preserved at the Veterinary Pharmacology Laboratory, University of Maiduguri, Nigeria.

Extract preparation

The root bark of the plant was collected, cleaned and then air-dried at room temperature for one week. It was pulverized using a mortar and pestle and 400 g of the ground herb was defatted by soaking it overnight in petroleum ether and then extracted by soaking it in 95% methanol for 24 hours. The resultant sample was filtered using Whatman filter paper No. 1 and evaporated to dryness under reduced pressure using a rotary evaporator R201D PEC Medicals.

Experimental animals

Forty-five albino rats of both sexes were procured from the animal house of Faculty of Veterinary Medicine, University of Maiduguri, Nigeria. They were kept in clean plastic rat cages in physiology research laboratory of the department of Veterinary physiology, University of Maiduguri, Nigeria and were fed with commercial feed and water *ad-libitum*. The animals were allowed to adjust to the laboratory environment for two weeks prior to commencement of the study.

Evaluation of anti-inflammatory effect of ethanolic extract of *Securidaca longipendunculata*

Five groups (A, B, C, D and E) of five rats each weighing between 100 g and 150 g were injected sub-plantally with 0.1 mL of fresh undiluted egg albumen to induce inflammation noting the time of induction [8]. Group A received the vehicle (distilled water) and served as the negative control, Group B, C and D received the crude methanol extract at a dose rate of 2, 4 and 8 mg/kg *i.p* respectively, while group E serving as the positive control received piroxicam (a known anti-inflammatory agent) at 10 mg/kg *i.p*. Thirty (30) minutes after the administration of egg albumin, inflammation was assessed in terms of change in diameter of the paw in all the groups at 1, 2, 3 and 4 hours after induction of inflammation. Fluid accumulation (edema) is associated with inflammation and the evaluation of anti-inflammatory activity was carried out by measuring the level of edema inhibition using the method described by Perez [9]. The percentage inhibition of edema was calculated, thus:

Percentage (%) inhibition = $\frac{(C_T - C_0)_{\text{control}} - (C_T - C_0)_{\text{treated}}}{(C_T - C_0)_{\text{control}}} \times 100$

Where, C_T = Mean paw diameter after egg albumin administration; C_0 = Mean paw diameter before egg albumin administration.

Sedative effect of methanolic extract of *Securidaca longipendunculata*

Four groups (A, B, C and D) of five rats each, weighing between 100 g and 150 g were used. Groups A, B and C received 2, 4 and 8 mg/kg (*i.p*) respectively of the crude methanol extract followed by the administration of diazepam (10 mg/kg) after thirty (30) minutes. Group D was also injected with normal saline (*i.p*) which served as the control animals followed by administration of diazepam thirty (30) minutes later. Sleeping time was assessed based on the length of time between sleep and wake using the loss and gain of righting reflex.

Statistical analysis

The values obtained were expressed as mean \pm standard deviation. Repeated measures analysis of variance was used to test significance in hourly values of the paw diameter. One way analysis of variance was used to test for significance in mean sleeping time among the treatments. $P \leq 0.05$ were considered significant. Graph pad InStat version 3.0 for windows USA computer software was used to analyze the data followed by Tukey-Kramer post hoc multiple comparison test.

Results

Anti-inflammatory effect of methanolic extract of *Securidaca longipendunculata* in albino rats

Effect of *S. longepedunculata* methanol extract on diameter of egg albumen induced paw edema in albino rats is presented in **Table 1**. Zero-hour paw diameter or pre-induction paw diameter was significantly lower than hourly (1, 2, 3 and 4) post induction diameters measured in negative control group. In all the extract treated groups, significant ($P < 0.05$) increase in paw diameter in first and second hour was observed. Group treated with standard drug (Piroxicam) showed significant increase from pre-induction value only at the first hour. This result indicated the extract has anti-inflammatory activity in the third hour onwards. In **Table 2**, at the first hour, the extract at 2 mg/kg, 4 mg/kg and 8 mg/kg provided 58.3%, 38.5% and 65.4% respectively when compared to piroxicam which conferred 69.2%. At the second hour, the extract conferred 47.1%, 29.4% and 64.7% protection in 2 mg/kg, 4 mg/kg and 8 mg/kg respectively, while piroxicam had 70.6%. At the third hour, the extract at 2 mg, 4 mg and 8 mg/kg conferred 94.4%, 82.2% and 88.8% protection respectively when compared with piroxicam which conferred 88.9%. At the fourth hour, the extract at 2 mg/kg, 4 mg/kg and 8 mg/kg conferred 90.9%, 81.8%

Table 1 Mean paw diameter of albino rats after *S. longepedunculata* methanolic extract administration.

Group (n=5)	Paw diameter (cm)				
	0 h	1 h	2 h	3 h	4 h
Negative control	4.1 \pm 0.22	6.7 \pm 0.44 ^a	5.8 \pm 0.57 ^a	5.9 \pm 0.22 ^a	5.2 \pm 0.27 ^a
2 mg/kg	4.2 \pm 0.31	5.4 \pm 0.36 ^a	5.1 \pm 0.4 ^a	4.3 \pm 0.27	4.3 \pm 0.11
4 mg/kg	4.3 \pm 0.27	5.9 \pm 0.41 ^a	5.5 \pm 0.35 ^a	4.62 \pm 0.23	4.1 \pm 0.44
8 mg/kg	3.9 \pm 0.12	4.8 \pm 0.32 ^a	4.5 \pm 0.15 ^a	4.1 \pm 0.17	3.9 \pm 0.16

a=Values in rows are significantly ($P < 0.05$) higher as compared to zero hour.

and 100% protection as compared to piroxicam which conferred 90.1% protection.

Sedative effect of methanolic extract of *Securidaca longepedunculata*

The results of sedative effect of *Securidaca longepedunculata* crude methanol extract in albino rats is presented in **Table 3**. There was no significant ($P < 0.05$) effect on the mean sleeping time of the test groups that were administered 2 mg/kg and 4 mg/kg when compared to the control group. In 8 mg/kg group, significant ($P < 0.05$) increase in mean sleeping time was observed when compared with the control group.

Discussion

Many plant extracts and synthetic products have been documented to possess anti-inflammatory [10] and analgesic [1] properties. In this study, *S. longepedunculata* root methanol extract was investigated for anti-inflammatory and central sedative effects via intra peritoneal routes. Diameter of induced paw edema reduced significantly at the third hour in 2, 4 and 8 mg/kg treated group which signified anti-inflammatory activity. Diazepam-induced sleeping time significantly ($P < 0.05$) increased in group treated with 8 mg/kg *S. longepedunculata* methanol extract which showed central sedating effect. Tissue inflammation involved edema formation due to increases in vascular permeability mediated by prostaglandins, leukotrienes, histamines and bradykinins release and inhibitors of these agents

Table 2 Percentage edema-inhibition of *S. longepedunculata* methanolic extract in albino rats.

Group	Edema inhibition (%)			
	1 st h	2 nd h	3 rd h	4 th h
2 mg/kg Extract	53.8	47.0	94.4	90.9
4 mg/kg Extract	38.4	29.4	83.3	81.8
8 mg/kg Extract	65.4	64.7	88.8	100
Piroxicam	69.2	29.4	12.5	90.9

Table 3 Effect of *S. longepedunculata* stem bark ethanolic extract on Mean sleeping time of diazepam-induced hypnosis.

Group	Sleeping time (s)
Diazepam (Control)	35 ± 19
2 mg/kg+Diazepam	27 ± 21
4 mg/kg+Diazepam	63 ± 14
8 mg/kg+Diazepam	88 ± 12 ^a

a=Value with superscript a in column is significantly ($P < 0.05$) different other values

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have anti-inflammatory properties [11]. Corticosteroids suppress inflammation by reducing the gene expression of these mediating substances which period of 24 hours to observe effect [12]. In this study, significant anti-inflammatory action was noticed in 3 hours and beyond signifying that the action could be due suppression of local re action which cause increased vascular permeability. The results of anti-inflammatory activity of *S. longepedunculata* agree with earlier studies reported [6]. Benzodiazepines stimulates inhibitory neurotransmitters, glycine and gamma amino butyric acid (GABA) in the central nervous system resulting in hypnosis [13]. In this study, sleeping time of was significantly ($P < 0.05$) increased in diazepam induced hypnosis. The central sedative activity of *S. longepedunculata* was also reported [5,6] and its anti-nociceptive activity was centrally mediated [14]. The analgesic activity of *S. longepedunculata* was more pronounced with methanol extracts given intraperitoneal [14] than aqueous extract following oral administration [5]. Similarly, the anti-inflammatory activity of the extract observed in this study was 88.8% at 8 mg/kg on the third hour which was more pronounced than the oral route (70% at 800 mg/kg) as Elufioye reported [15]. It also increased sleeping time induced by diazepam by 53 seconds given at 10 mg/kg and when compared with result were obtained [5] prolongation of sleep was by 65 seconds by Hexobarbitone induced sleep. The higher activity observed by Adeyemi et al may be ascribed to the difference in the experimental model in which a more potent drug (Hexobarbitone) was used to induce sleep. The conventional analgesic drugs include non-steroidal anti-inflammatory drug (NSAID) and narcotic drugs [16]. Collateral anti-inflammatory effect is reported in both classes of drugs [17-19]. In addition, central sedating activity is reported in narcotic analgesic which NSAID lacked [20]. Opioids potentiate the hypnotic activity of benzodiazepines [21]. The prolongation of diazepam induced sleeping time may signify similar pharmacodynamic action of the extract with opioids. Outcome of this study shows anti-inflammatory and central sedating activity of *S. longepedunculata* methanolic extract, work should be carried out elucidate the mechanism of these activities.

Conclusion

Methanol extract of *S. longepedunculata* root possess anti-inflammatory with sedative properties. This justifies the reported efficacy in traditional use of the plant for the same purpose and can be adopted as an alternative therapy in the management of various inflammatory conditions. Further research to identify and characterize the specific compound(s) responsible for these effects is recommended.

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