

DOI: <https://dx.doi.org/10.18203/2319-2003.ijbcp20212066>

Original Research Article

Anti-nociceptive, anti-inflammatory and antipyretic activities of the ethanol root bark extract of *Salacia lehmbachii* in rats and mice

Godwin C. Akuodor^{1*}, Sylvester C. Ohadoma², Casimir C. Ofor³, Anthony U. Megwas⁴, Leo C. Chukwu⁵, Mansur A. Ramalan⁶, Dorcas O. Okoroafor⁷, Kingsley C. Chilaka¹

¹Department of Pharmacology and Therapeutics, Faculty of Medicine, College of Health Sciences, Nnamdi Azikiwe University, Nnewi Campus, Nigeria

²Department of Pharmacology, College of Medical Sciences, University of Calabar, Nigeria

³Department of Pharmacology and Therapeutics, Faculty of Medicine, Ebonyi State University, Abakaliki, Nigeria

⁴Department of Optometry, School of Health Technology, Federal University of Technology, Owerri, Nigeria

⁵College of Medicine, Chukwuemeka Odumegwu Ojukwu University, Amaku, Awka, Nigeria

⁶Department of Pharmacology and Therapeutics, College of Health Sciences, Bayero University, Kano

⁷Department of Biomedical Technology, School of Science Laboratory Technology, University of Port Harcourt, Nigeria

Received: 21 February 2021

Accepted: 31 April 2021

*Correspondence:

Dr. Godwin C. Akuodor,

Email: goddyakuodor@yahoo.com

Copyright: © the author(s), publisher and licensee Medip Academy. This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial License, which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

ABSTRACT

Background: The decoction of the roots of *Salacia lehmbachi* is used in traditional medicine for the treatment of different diseases such as malaria, diabetes and microbial infections.

Methods: Phytochemical screening and oral acute toxicity tests were carried out on the ethanol root extract of the plant. Anti-nociceptive activity using acetic acid induced writhing and tail immersion method in mice, anti-inflammatory activity using carrageenan induced paw oedema in rats and xylene induced ear oedema test in mice and antipyretic activity using Brewer's yeast and D-amphetamine induced pyrexia in rats were determined at 50 mg/kg, 100 mg/kg and 200 mg/kg doses of the root extract.

Results: The ethanol root extract contains alkaloids, saponins, tannins, flavonoids, terpenoids, steroids and cardiac glycosides. The oral acute toxicity tests were found to be greater than 5000 mg/kg. The root extract and the standard drug (Aspirin) significantly ($p < 0.05$ and $p < 0.01$) decreased the number of writhes caused by acetic acid. The extract and morphine significantly ($p < 0.05$ and $p < 0.01$) prolonged reaction time in tail immersion model. The extract produced significant ($p < 0.05$ and $p < 0.01$) dose dependent inhibition of oedema which was comparable to aspirin in carrageenan induced paw oedema model. The root extract also demonstrated significant ($p < 0.05$ and $p < 0.01$) effect in xylene induced mouse ear oedema test compared to dexamethasone. The extract significantly decreased high temperature in both Brewer's yeast and d-amphetamine induced pyrexia.

Conclusions: Findings show that *S. lehmbachii* may provide a good source of plant compounds with analgesic, anti-inflammatory and antipyretic activities.

Keywords: *Salacia lehmbachii*, Analgesic, Anti-inflammatory, Antipyretic, Rats, Mice

INTRODUCTION

Inflammation which is a response to infection or injury is characterized by pain, heat, swelling, redness and

disrupted physiological functions. Chemical mediators released from injured tissue and migrating cells can induce inflammation.¹ Non-steroidal anti-inflammatory drugs (NSAIDs), aspirin and opiates generally used for the

management of inflammatory conditions have not been successful in all cases as a result of adverse effects such as liver damage and ulcers.^{2,3} The strategy should therefore be the search for clinically new and useful anti-inflammatory agents which have negligible or no side effects. It has been recorded that agents derived from natural products are able to affect a wide range of pathways involved in the inflammatory response, including different inflammatory mediators like cytokines, arachidonic acid metabolites, excitatory amino acids, peptides and others.¹ Fever is due to the elevation of core body temperature above normal.⁴ Fever may be due to infection, inflammation, or any tissue damage and disease state. It arises as a secondary impact of infection, malignancy or other diseased states.⁵

Salacia lehmbackii Loes which belongs to the family Celastraceae is used in traditional medicine for different ailments. This plant is native to Nigeria, Cameroon, Liberia, Senegal and Tanzania. Different studies showed the antidiarrhoeal, antimicrobial and anti-ulcer effects of both the leaves and the root of the plant.^{6,7} More so, anticholinergic activity, anti-infertility and anti-hemorrhoid effects, as well as antioxidant activity were studied.⁸⁻¹¹ The objective of this study was to evaluate the anti-nociceptive, anti-inflammatory and antipyretic activities of ethanol extract of *S. lehmbackii* root bark in rats and mice.

METHODS

Plant collection

The roots of *Salacia lehmbackii* were harvested from a farm land at Ukanafun local government area, Akwa Ibom State, Nigeria. The plant was identified and authenticated in the herbarium of the department of botany, University of Calabar. The specimen with registration number 688 was deposited at botany department herbarium, University of Calabar.

The roots were collected air-dried for fourteen days and the size was reduced with mortar and pestle. The powdered material was then subjected to extraction using soxhlet extractor for 48 hours, and the filtrate was evaporated on water bath at reduced temperature of 45°C to obtain a brown solid residue.

Experimental animals

Male and female wistar rats and mice weighing 150-180 g and 20-25 g respectively were obtained from the animal house of the department of pharmacology and therapeutics, University of Calabar. The animals were allowed free access to standard feed and water *ad libitum*. They were kept six each in clean separate cages with saw dust as bedding, which was replaced every two days.

The study was conducted according to ethical guidelines on laboratory animal use and care which is in compliance with University of Calabar Research Policy

(ERN/025PA30617) and the authors of this manuscript observed ethical issues. The National Institute of Health Guide for the care and use of laboratory animals was also adopted for this study.¹²

Acute toxicity test

Acute toxicity testing of ethanol root bark of *S. lehmbackii* extract was estimated in both rats and mice using modified method in two phases.¹³ The animals were deprived of food overnight prior to administration of the extract. In phase 1, three groups of three animals in each cage were used. The root bark extract was orally administered geometrically in increasing doses of 10 mg/kg, 100 mg/kg and 1000 mg/kg respectively. The animals were then monitored for first 4 hours for signs of toxicity and mortality for 24 hours. When no lethality was observed, phase 2 was introduced. In second phase, three groups of one animal were intragastrically given the extract in geometrically increasing doses of 1600 mg/kg, 2900 mg/kg and 5000 mg/kg respectively. The animals were also observed for first 4 hour for physical signs of toxicity and mortality for 24 hours.

Anti-nociceptive test

Acetic acid induced writhing in mice

The ethanol root bark extract was assessed for analgesic activity using acetic acid-induced writhing method.¹⁴ Albino mice (20-25 g) of both sexes randomized into 5 different cages of 6 mice in each cage. Group 1 (drug free) was given normal saline (20 ml/kg), the extract (50 mg/kg, 100 mg/kg and 200 mg/kg) were administered to group 2-4, respectively. The positive control received (150 mg/kg) of acetyl salicylic acid (ASA).

After thirty minutes, intraperitoneal injection of acetic acid (10 ml/kg, 0.7%) was administered to each mouse. They were separately placed in a transparent cage for observation. The writhing movements for each mouse was counted for 30 minutes.

Tail immersion test

Pain latencies of the extract was measured according to the methods.^{15,16} Albino mice of both sexes grouped into 5 cages with 6 mice per group were assayed. Group 1 was treated with normal saline, groups 2-4 were treated with different doses of the extracts (50 mg/kg, 100 mg/kg and 200 mg/kg). The last group 5 was treated with morphine (10 mg/kg). Thirty minutes after treatment, each mouse was kept in a restrainer cage with the tail freely hanging inside hot water of 50±1°C temperature.

The results were recorded after tail withdrawal. Readings were taken at 30, 60, 90 and 120 minutes interval. The mice used were initially screened and those that did not attempt to withdraw the tail in 10 seconds were not listed for the study.

Anti-inflammatory effect

Carrageenan induced paw oedema test

The carrageenan-induced paw oedema test as described by was used with slight modification.¹⁷ Thirty wistar rats grouped into five with six rats in each cage were used for the experiment. Group 1 served as negative control (10 mL/kg distilled water), group 2 positive control (150 mg/kg aspirin), while groups 3, 4 and 5 were administered 50 mg/kg, 100 mg/kg and 200 mg/kg of the ethanol root bark extract of *S. lehmbachii* respectively. Acute inflammation was induced 30 min after treatment by administration of 0.1 mL of 1% suspension of carrageenan into the subplantar right hind paw of the rats. Assessment of volume of oedema using plethysmometer was implemented. Reading was first taken before treatment and after 20 min intervals for 2 hours.¹⁸

Xylene induced ear oedema method

This method as described by was adopted but with slight modification.¹⁹ Mice were grouped into 5 with 6 animals in each. The animals were orally treated with the root bark extract in graded doses of 50 mg/kg, 100 mg/kg and 200 mg/kg. The negative control group was treated with 10 ml/kg of distilled water, while the positive control was treated with 4 mg/kg of dexamethasone. Oedema was induced in each mouse one hour after treatment with a drop xylene into the inner surface of the right ear. Three hour later, mice were sacrificed and both cut-off to equal size and weighed. The mean difference between the right and left ears were recorded as an indication of inflammation.

Antipyretic activity

Brewer's yeast induced pyrexia

The antipyretic activity was evaluated using Brewer's yeast induced pyrexia in rats as described.²⁰ Fever was induced by administering 20 ml/kg of 20% aqueous suspension of Brewer's yeast in distilled water subcutaneously 24 hours before treatment. Thirty wistar rats of both sexes were divided into five groups, 1 and 2 served as negative control (distilled water 20 ml/kg) and positive control (paracetamol 150 mg/kg), while groups 3, 4, and 5 received 50 mg/kg, 100 mg/kg, and 200 mg/kg of the extract respectively. All drugs were administered orally. Rectal temperatures were taken by the use of digital thermometer (Mediklin, China) before yeast injection, 24 h after the injection, and at 1, 2, 3, 4 and 5 hours after drug administration.

Amphetamine induced pyrexia in mice

The study was in accordance to Okokon et al protocol, with little modification.²¹ The initial temperatures of rats recruited for this study were taken. The animals were grouped and each rat was administered with 5 mg/g of D-amphetamine intraperitoneally to induce pyrexia. Twenty-four hours after, the rectal temperature of each rat was

checked to confirm increase in temperature and animals showing increase in temperature of less than 0.6°C were not recruited for the study. Thereafter, animals were treated with graded doses of the root extract (50 mg/kg, 100 mg/kg and 200 mg/kg) were administered orally to three groups of wistar rat, while distilled water (20 ml/kg) and 150 mg/kg of paracetamol were administered to the remaining two groups (drug free and standard drug). The rectal temperature of each rat was then observed at interval of 1 hour for 5 hours.

Statistical analysis

Results are presented as mean±standard error of mean (SEM) and analyzed with statistical package for social sciences (SPSS version 20) using one-way analysis of variance (ANOVA) followed by Dunnett's post hoc test. Difference in the mean ($p < 0.05$) was statistically considered significant.

RESULTS

Phytochemical analysis

The phytochemical screening of *S. lehmbachii* ethanol root extract revealed the presence of alkaloids, saponins, tannins, flavonoids, terpenoids, steroids, cardiac glycosides, resins and balsam as reported by Akuodor et al.²²

Acute toxicity studies

There was no observed changes, mortality or signs of toxicity 72 hours after administration of the ethanol root extract. The animals were all healthy and active throughout the study. Hence, the median lethal dose (LD₅₀) was found to be greater than 5000 mg/kg.

Anti-nociceptive studies

The root extract of *S. lehmbachii* significantly and dose dependently decreased the number of acetic-acid induced writhes at $p < 0.05$ and $p < 0.01$. The observed effects of the extract at 200 mg/kg was higher than that of 50 mg/kg and 100 mg/kg. This effect was comparable to that of the standard drug (Table 1).

Table 1: Effect of ethanol extract of *S. lehmbachii* root bark on acetic acid-induced writhing in mice.

Treatment	Dose (mg/kg)	Mean no. of writhes	Inhibition (%)
Distilled water	20 ml/kg	23.33±4.93	-
<i>S. lehmbachii</i>	50	9.00±1.38	61 ^a
	100	5.33±1.11	77 ^a
	200	3.50±0.72	85 ^b
Aspirin	150	3.00±0.52	87 ^b

Results are mean±SEM; (n=6), ^aP<0.05, ^bP<0.01 compared to control.

Table 2: Effect of ethanol extract of *S. lehmbachii* root bark on tail immersion in mice time interval (min).

Treatment	Dose mg/kg	Pre-treatment		After-treatment		
		0	30	60	90	120
<i>S. lehmbachii</i>	50	6.50±0.43	8.50±0.50	10.83±0.83	11.67±0.71	13.17±0.60 ^a
	100	7.50±0.76	9.33±0.76	11.83±0.70	13.50±0.56	15.00±0.68 ^a
	200	8.00±0.52	10.17±0.79	12.50±0.88	14.67±0.56	17.50±0.34 ^b
Morphine	10	7.50±0.62	17.50±0.72	19.17±0.60	21.17±0.87	23.33±0.95 ^b

Results are mean±SEM; (n=6) ^aP<0.05, ^bP<0.01 compared to control

Table 3: Effect of ethanol extract of *S. lehmbachii* root bark on carrageenan induced paw oedema in rats.

Treatment	Dose (mg/kg)	Paw oedema (hours)						
		0	0.5	1	2	3	4	5
Distilled water	20 ml/kg	1.24±0.02	1.64±0.01	1.72±0.02	1.79±0.02	1.86±0.02	1.93±0.0	2.01±0.02
<i>S. lehmbachii</i>	50	1.22±0.03	1.60±0.02	1.53±0.03	1.46±0.02	1.38±0.03	1.31±0.02*	1.20±0.02 ^a
	100	1.24±0.02	1.63±0.01	1.56±0.02	1.48±0.02	1.30±0.03	1.24±0.02	1.18±0.02 ^a
	200	1.18±0.03	1.57±0.03	1.49±0.03	1.38±0.02	1.30±0.04	1.21±0.03	1.15±0.03 ^b
Aspirin	150	1.17±0.04	1.61±0.02	1.49±0.02	1.39±0.03	1.30±0.03	1.22±0.03	1.12±0.03 ^b

*Results are mean± SEM; (N=6) ^aP<0.05, ^bP<0.01 compared to control

Table 4: Effect of ethanol extract of *S. lehmbachii* root bark on xylene-induced ear oedema in mice.

Treatment	Dose (mg/kg)	Weight of ear			Inhibition (%)
		Right ear	Left ear	Increase in weight	
Distilled water	20 ml/kg	0.065±0.00	0.300±0.00	0.020±0.00	-
<i>S. lehmbachii</i>	50	0.052±0.00	0.030±0.00	0.020±0.00	51 ^a
	100	0.040±0.00	0.028±0.00	0.016±0.00	63 ^a
	200	0.030±0.00	0.018±0.00	0.008±0.00	83 ^b
Dexamethazone	4	0.025±0.00	0.015±0.00	0.007±0.00	85 ^b

Results are mean± SEM; (N=6) ^aP<0.05, ^bP<0.01 compared to control

The ethanol root extract of *S. lehmbachii* significantly at p<0.05 and p<0.01) reduced the thermal stimuli in mice. In this test, dose dependent reduction was produced by the extract. However, morphine (reference drug), showed stronger protection (Table 2).

Anti-inflammatory studies

The anti-inflammatory activity of *S. lehmbachii* ethanol root extract was observed to be dose dependent. There was significant activity at p<0.05 with 50 mg/kg and 100 mg/kg doses of the extract, whereas the highest dose of the extract (200 mg/kg) had p<0.01 significant activity. The reference drug (ASA) had more activity in this study (Table 3). The anti-inflammatory activity began at 1 hour after administration of drug and lasted for 5 hours before completely vanishing.

The anti-inflammatory effect of *S. lehmbachii* ethanol root extract against xylene induced ear oedema in mice is

shown in Table 4. The extract exhibited significant and dose dependent activity reduction of oedema at p<0.05 and p<0.01 with the highest dose of the extract comparable to the standard drug, dexamethasone.

Antipyretic studies

Table 5 shows the antipyretic effect of the ethanol root extract of *S. lehmbachii* determined using Brewer’s yeast induced pyrexia in rats. The extract exerted significant and dose dependent antipyretic action at p<0.05, whereas the standard drug showed significant antipyretic activity at p<0.01.

The results of the effect of ethanol root bark extract of *S. lehmbachii* against D-amphetamine induced pyrexia is shown in Table 6. There was a progressive dose dependent reduction at p<0.05 in the temperature of rats treated with the ethanol extract. The effect of the extract was less than the standard drug, acetylsalicylic acid (ASA).

Table 5: Effect of ethanol extract of *S. eahmbachii* root bark against yeast induced pyrexia in rats (hours).

Treat-ment	Dose (mg/kg)	0	24	1	2	3	4	5
Distilled water								
	20 ml/kg	35.37±0.05	37.52±0.04	37.80±0.02	37.63±0.02	37.42±0.02	37.29±0.02	37.30±0.03
<i>S. lehmbachii</i>								
	50	35.26±0.03	37.25±0.03	36.41±0.01	36.20±0.02	35.62±0.03	35.37±0.03	35.24±0.02 ^a
	100	35.25±0.02	37.27±0.02	36.50±0.03	36.22±0.01	35.61±0.02	35.36±0.03	35.22±0.03 ^a
	200	35.23±0.02	37.30±0.02	36.47±0.02	36.15±0.01	35.55±0.03	35.30±0.02	35.10±0.03 ^a
Aspirin								
	150	35.22±0.00	36.79±0.02	35.69±0.04	35.43±0.02	35.40±0.01	34.60±0.02	34.30±0.03 ^b

Results are mean±SEM; (N=6) ^aP<0.05, ^bP<0.01 compared to control.

Table 6: Effect of ethanol root bark extract of *S. lehmbachii* on de-amphetamine induced pyrexia in rats (hours).

Treatment	Dose (mg/kg)	0	24	1	2	3	4	5
Distilled water								
	20 ml	35.25±0.04	37.39±0.04	37.61±0.04	37.67±0.02	37.46±0.05	37.26±0.03	37.70±0.03
	50	35.27±0.04	37.29±0.02	36.43±0.03	36.23±0.03	35.51±0.02	35.30±0.02	35.25±0.02 ^a
<i>S. lehmbachii</i>	100	35.20±0.03	37.30±0.02	36.40±0.02	36.23±0.02	35.52±0.02	35.31±0.02	35.23±0.03 ^a
	200	35.24±0.02	37.30±0.02	36.33±0.02	36.20±0.03	35.48±0.02	35.26±0.02	35.21±0.01 ^a
Paracetamol	150	35.25±0.03	37.31±0.02	36.39±0.03	36.22±0.03	35.49±0.01	35.31±0.01	34.17±0.01 ^a

Results are mean±SEM; (N=6) ^aP<0.05, ^bP<0.01 compared to control

DISCUSSION

This study was carried out to establish the potential pharmacological properties of ethanol extract of *S. lehmbachii* based on claims of its use in herbal medicine. The findings of the present study reveal that *S. lehmbachii* (ethanol extract) at doses employed exhibited anti-nociceptive effects against chemical pains (writhing) induced by acetic acid.²³ This chemical is popular for evaluating anti-nociceptive properties of plant extracts and drugs.²⁴ Another report revealed that the response of mice to acetic acid is a fast and trusted method to test peripheral anti-nociceptive effect of herbal drugs.²⁵

In the present work, the extracts of *S. lehmbachii* and acetylsalicylic acid (aspirin) inhibited acetic acid induced writhing. The results shows the extract has peripheral anti-nociceptive properties which suggest the action may be directed via inhibition of local peritoneal receptors.²⁶ However, irrespective of whether the model evaluates peripheral anti-nociceptive action only or non-specific, the results validates the usefulness of this plant as an analgesic in Nigeria. The injection of acetic acid is reported to induce the release of mediators of pain such as prostaglandins and other cyclokinase.^{27,28} This suggests that the extracts of *S. lehmbachii* acted by inhibiting the actions of cyclooxygenase which is said to be responsible for producing prostaglandins from arachidonic acids.²⁸ The analgesic effects produced by the extract of the test plant (*S. lehmbachii* root bark) validate their use in traditional medical practice as analgesics.

In order to confirm the analgesic activity of the extract, centrally acting model of analgesia (tail immersion) was carried out. This method of analgesic assay which is used to indicate the involvement of central analgesic mechanism is believed to involve spinal reflex.²⁹ It has been reported that centrally acting agents such as morphine, possess this activity in both types of study, whereas peripherally acting agents lie acetylsalicylic acid have been reported to exert anti-nociceptive action only in the writhing test.³⁰ Most importantly, the action of acetylsalicylic acid in writhing assay only could be linked with its ability to directly inhibit prostaglandin activity or indirectly inhibit prostaglandin secretion by inhibition of cyclo-oxygenase activity.³¹ The importance activity in tail immersion test shows involvement of central analgesic mechanism.

Carrageenan induced paw oedema is a suitable way of testing anti-inflammatory agents and this has widely been used for screening anti-oedematous action of natural products.³² This process of testing acute inflammation potency is a highly sensitive tool.³³ The development of oedema depend on the presence of bradykinin and polymorphonuclear leucocytes with pro-inflammatory factor such as prostaglandins.³⁴ The root extract of *S. lehmbachii* may not have activity on the early phase of inflammation, hence may act by inhibiting the release of prostaglandins. Nonsteroidal anti-inflammatory agents like aspirin, may not inhibit the initial phase of edema induced by carrageenan whereas the second accelerating phase can be antagonized by the drug.³⁵

Xylene causes irritation in mouse ear leading to accumulation of fluid and oedema and increase in myeloperoxidase enzymatic activity.³⁶ Suppression of this response may suggest antiphlogistic activity.²⁹ The ethanol extract of *S. lehmbachii* root exerted significant inhibition of ear oedema in mice. This activity suggests the inhibition of phospholipase A₂ which is involved in the pathophysiology of inflammation due to xylene.³⁷ However, dexamethasone used as the reference drug exhibited significant decrease in the ear weight of positive control rats which indicate an inhibition of PLA₂.

Antipyretic agents have been shown to antagonize cyclooxygenase activity via increase in prostaglandin E₂ thereby suppressing high temperature.³⁸ Rise in temperature may be caused by damaged tissue, infections, and other diseases. This process will give rise to mediators (interleukins etc.), which progresses to prostaglandin E₂ formation with increase body temperature.³⁹ The extracts reduced rats' anal temperature. The observed effect was similar to that of aspirin. The ethanol root extract of *S. lehmbachii* could bring fever to a control by getting rid of inflammatory symptoms at both peripheral and nervous system thermoregulator zones. They could bring down pyrogenic secreting cytokines while reducing prostaglandin E₂ synthesis from cyclooxygenase probably through the mechanism reputed for paracetamol.⁴⁰

The therapeutic potentials of medicinal plants are mostly attributed to the combination of secondary metabolites. Flavonoids have been reported to target prostaglandins involved in late phase of acute inflammation and pain and they have therefore been associated with anti-nociceptive, anti-inflammatory and antipyretic activities.⁴¹⁻⁴³ Therefore, it is not surprising to have seen these activities in the root extract. The LD₅₀ result obtained showed the relative safety of the herbal agent as no death was recorded.

CONCLUSION

The results of present study show that the ethanol root of *S. lehmbachii* possess anti-nociceptive, anti-inflammatory and antipyretic activities, thus confirming the folklore uses of the plant for the treatment of different diseases.

ACKNOWLEDGEMENTS

Authors are grateful to Mr. Marcus Inyang and Etim Ifang for their technical assistance.

Funding: No funding sources

Conflict of interest: None declared

Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Chaudhari MG, Joshi BB, Mistry KN. In vitro anti-diabetic and anti-inflammatory activity of stem bark of

- Bauhinia purpurea*. Bull Pharm Med Sci. 2013;1(2):139-50.
2. Zulfiker AHM, Rahman MM, Hossain MK, Hamid K, Mazumder MEH, Rana MS. In vivo analgesic activity of ethanolic extracts of two medicinal plants- *Scoparia dulcis* L. and *Ficus racemosa* Linn. Biol Med. 2010;2(2):42-8.
 3. Bellik Y, Boukraa L, Alzahrani HA, Bakhotmah BA, Abdallah F, Hammoudi SM, et al. Molecular mechanism underlying anti-inflammatory and anti-allergic activities of phytochemicals: An update. Molecules. 2012;18:322-53.
 4. Dalal S, Zhukovsky DS. Pathophysiology and management of fever. J Support Oncol. 2006;4:9-16.
 5. Tirumalasetty J, Ubedulla S, Chandrasekhar N, Kishan PV, Rasamal K. Evaluation of antipyretic activity of alcoholic extract of *Vitex nigundo* leaves in PGE1 induced pyrexia model in Albino Rats. J Chem Pharm Res. 2012;4(6):3015-19.
 6. Essien AD, Akuodor GC, Essien EA, Asika EC, Chilaka KC, Nwadam SK. Evaluation of antipyretic potential of the ethanolic leaf extract of *Salacia lehmbachii* Loes. Asian J Med Sci. 2015;7(2):22-5.
 7. Essien AD, Takem LP, Anele EI. In vitro cholinergic and acute toxicity evaluations of *Salacia lehmbachii*. Int J Pharm Pharm Res. 2016;5(1):200-7.
 8. Takem LP, Lawal BAS, Udia PM. Analgesic and acute anti-inflammatory activities of aqueous root extract of *Salacia lehmbachii*. Br J Pharm Res. 2014;4(18):2172-81.
 9. Essiet GA, Essien AD, Udoh FV, Essiet A. Anti-fertility effects of ethanol extract of *Salacia lehmbachii* root bark in albino rats. J Adv Med Pharm Sci. 2016;8(4):1-8.
 10. Sofowora A. Medicinal Plants and Traditional Medicine in Africa. Ibadan: Spectrum Books; 1993: 200.
 11. Akuodor GC, Essiet GA, Essien AD, Udoh FV, Ogiji DE, Nwadam SK, et al. In vitro antioxidant activity of *Salacia lehmbachii* ethanol root bark extract. Eur J Med Plants. 2017;18(4):1-6.
 12. National Institutes of Health. Guide for the Care and Use of Laboratory Animals. 8th ed. Bethesda, MD: NIH; 2011. 82-3.
 13. OECD. OECD Guideline for Testing of Chemicals (TG 401). Acute Oral Toxicity- Fixed Dose Procedure. 2011.
 14. Koster R, Anderson M, Beer EJ. Acetic acid analgesic screening. Federation Proc. 1959;18: 412-7.
 15. Akuodor GC, Anyalewechi NA, Udoh FV, Ikoro NC, Akpan JL, Gwotmut MD, Pharmacological evaluation of *Verbena hastate* leaf extract in the relief of fever. Adv Pharmacol Toxicol. 2011;12(3):1-8.
 16. Kalpesh G, Nema RK, Kori ML, Sharma CS, Singh V. Anti-inflammatory and analgesic activity of *Balanites aegyptiaca* in experimental animal models. Int J Gastroenterol. 2008;214-7.
 17. Winter CA, Risley EA, Nuss GW. Carrageenin-induced edema in hind paws of the rat as an assay for

- anti-inflammatory drugs. *Proc Soc Exp Med.* 1962;111:544-7.
18. Oyewole IO, Ibidapo CA, Moronkola DO, Oduola AO, Adeoye GO, Anyasor GN, et al. Antimalarial and repellent activities of *Tlthonia diversifolia* (Hemsl.) leaf extracts. *J Med Plants Res.* 2008;2(8):171-5.
 19. Agbaje EO, Ajidahun OA. Analgesic, Anti-inflammatory and antipyretic effect of dried root ethanolic extract of *Strophantus sarmentosus* (apocynaceae). *Int Res J Pharm Pharmacol.* 2011;62-9.
 20. Hassan FI, Abdulkadir UZ, Yaro AH, Danmalam UH. Analgesic, anti-inflammatory and antipyretic activities of the methanol leaf extract of *Dalbergia saxatilis* Hook. in rats and mice. *J Ethnopharmacology.* 2015;166:74-8.
 21. Akuodor GC, Essiet GA, Ekenjoku JA, Udoh FV, Ogiji ED, Ibiam GA, et al. Antidiarrhoeal and antimicrobial effects of ethanol root bark extract from *Salacia lehmbachii*. *J Herbmed Pharmacol.* 2021;10(2):188-93.
 22. Okokon JE, Nwafor PA. Anti-inflammatory, analgesic and antipyretic activities of ethanolic root extract of *Croton zambesicus*. *Pak J Pharm Sci.* 2010;23(4):385-92.
 23. Shaa KK, Oguche S, Watila IM, Ikpa TF. In vitro antimalarial activity of the extracts of *Vernonia amygdalina* commonly used in traditional medicine in Nigeria. *Science World J.* 2011;6(2):5-9.
 24. Du J, Yu Y, Ke Y, Wang C, Qian ZM. Ligustilide attenuates pain behavior induced by acetic acid or formalin. *J Ethnopharmacology.* 2007;112:211-4.
 25. Essien AD, Edidara Thomas, Essiet GA, Akuodor GC. Anti-inflammatory, antipyretic and anti-nociceptive activities of the ethanol stem bark extract of *Salacia lehmbachii*. *British Pharmacol Toxicol.* 2017; 8(2):9-16.
 26. Mbiancha M, Kamanyi A, Teponno RB, Taponjoun AL, Watcho P, Nguelefack T. Analgesic and Anti-Inflammatory Properties of Extracts from the Bulbils of *Dioscorea bulbifera* L. var *Sativa* (Dioscoreaceae) in Mice and Rats. *Evid Based Compl Altern Med.* 2011:912935.
 27. Divya TS, Latha PG, Usha K, Anuja GI, Suja SR, Shyamal S, et al. Anti-inflammatory, analgesic and anti-lipid peroxidative properties of *Wattakaka volubilis* (Linn). *Natur Prod Rad.* 2009;8(2):137-41.
 28. Nkeh CNB, Bekwa PCM, Ndebia JE, Kayo M, Mbafor TJ, Iputo EJ. Analgesic and anti-inflammatory properties of *Oxyanthus unilocularis*. *J Med Plants.* 2010;4(10):932-9.
 29. Akuodor GC, Essien AD, Udia PM, David-Oku E, Chilaka KC, Asika EC, Nwadam SK. Analgesic, Anti-inflammatory and Antipyretic potential of the stem Bark Extract of *Stachytarpheta indica*. *British J Pharmacol Toxicol.* 2015;6(1):16-21.
 30. Ezeja M, Ezeigbo I, Madubuike KG. Analgesic activity of the methanolic seed extract of *Buchholzia coriacea*. *Res J Pharm Biol Chem Sci.* 2011;2(1):187-93.
 31. Kumar A, Agarwal K, Kumar MA, Shanker K, Bushra U, Tandon S, et al. Pharmacological and phytochemical evaluation of *Ocimum sanctum* root extract for its anti-inflammatory, analgesic and antipyretic activities. *Pharmacognosy Mag.* 2015;11(42):217-24.
 32. Panthong A, Kanjanapothi D, Taesotikul T, Wongcome T, Reutrakul V. Anti-inflammatory and antipyretic properties of *Clerodendrum petasites*. *J Ethnopharmacology.* 2003;58:151-6.
 33. Xu Z, Zhou J, Cai J, Zhu Z, Sun X, Jiang C. Anti-inflammation effects of hydrogen saline in LPS activated macrophages and carrageenan induced paw oedema. *Inflammation.* 2012;9:2.
 34. Necas J, Bartosikova L. Carrageenan: A review. *Veterinary Med.* 2013;58:187-205.
 35. David OE, Akuodor GC, Edet EE, Ogbuji GK, Obiajunwa OJI, Aja DOJ. Antinociceptive, anti-inflammatory and antipyretic effects of ethanolic root bark extract of *Ipacina senegalensis* in rodents. *J Appl Pharmaceut Sci.* 2016;6(02):104-3.
 36. Yazmin RC, Vivian MC, Yohani PG, Ambar OY, Sonia JD, Rosa MF. Anti-Oedema Effects of D-002 and Lyprinol on the Carrageenan-Induced Pleurisy in Rats. *Int J Pharm Sci Rev Res.* 2013;23(1):24-8.
 37. Megwas AU, Akuodor GC, Chukwu LC, Aja DO, Okorie EM, Ogbuagu EC, et al. Analgesic, anti-inflammatory and antipyretic activities of ethanolic extract of *Annona senegalensi* leaves in experimental animal models. *Int J Basic Clin Pharmacol.* 2020;9(10):1477-84.
 38. David M, Aronoff MD, Eric G, Neilson MD. Antipyretics: mechanisms of action and clinical use in fever suppression. *Amer J Med.* 2001;111(4):304-15.
 39. Boron WF, Boulpaep EL. *Medical Physiology: A Cellular and Molecular Approach.* 2nd ed. Philadelphia, PA : Saunders/Elsevier, 2009; 1300.
 40. Sumanta M, Gouri KD, Suman A. Analgesic, anti-inflammatory and antipyretic studies of *Neolamarckia cadamba* barks. *J Pharma Res.* 2009;1:1133-6.
 41. Agoreyo BO, Okoro NC, Choudhary MI. Preliminary phytochemical analysis of two varieties of *Adenia lobate* and the antioxidant activity their various solvent fractions. *Bayero J Pure and Appl Sci.* 2012;5(1):182-6.
 42. Savithamma M, Rao ML, Suvrulatha D. Screening of medicinal Plants for secondary metabolites. *Middle-East J Scient Res.* 2011;8(3):579-84.
 43. Nassar N, Abeywardana P, Barker A, Bower C. Parental occupational exposure to potential endocrine disrupting chemicals and risk of hypospadias in infants. *Occupational. Environment Medicine.* 2010;67:585-9.

Cite this article as: Akuodor GC, Ohadoma SC, Ofor CC, Megwas AU, Chukwu LC, Ramalan MA., et al. Anti-nociceptive, anti-inflammatory and antipyretic activities of the ethanol root bark extract of *Salacia lehmbachii* in rats and mice. *Int J Basic Clin Pharmacol* 2021;10:614-20.