

# ANTI-INFLAMMATORY AND TOXICITY PROPERTIES OF METHANOL ROOT EXTRACT OF *SARCOCEPHALUS LATIFOLIUS* IN ALBINO RATS

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**Abstract:** Medicinal plants have the potential to provide compounds of novel and complex structures that are capable of interacting with biological systems. The research into plants with medicinal use as anti-inflammatory and pain relievers is considered as a fruitful and logical strategy in the search for a new anti-inflammatory and analgesic drugs. Although a large number of synthetic and clinically useful anti-inflammatory and analgesic drugs are available in the market, the search for new effective drugs with meaningful safety profile remains vital. Adult albino rats of both sexes weighing between 150-200g/w were used for the experiment and the result obtained showed that the methanol root extract of *S. Latifolius* significantly reduced inflammation. The extract was found to inhibit carrageen induced paw oedema significantly ( $P < 0.05$ ) by inhibiting mediators of inflammation. This suggests that the extract possess both anti-inflammatory and analgesic activity that might be mediated through a common mechanism. Similarly the toxicity profile of the plant extract was assayed and the result obtained showed that there are no signs of toxicity at the acute level but long exposure can pose a threat to healthy living as revealed by the result of the sub-chronic toxicity. It is therefore established that the methanol extract of *S. Latifolius* has anti-inflammatory activity.

**Keywords:** Analgesic, Methanol, Root, screening, Activity.

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## 1. INTRODUCTION

Medicinal plants have the potential to provide compounds of novel and complex structures that are capable of interacting with biological systems (Hostettmann *et al.*, 2005). The research into plants with medicinal use as anti-inflammatory and pain relievers should therefore be considered as a fruitful and logical strategy in the search for a new anti-inflammatory and analgesic drugs (Macía, 2005). Although a large number of synthetic and clinically useful anti-inflammatory and analgesic drugs are available in the market, the search for new effective drugs with meaningful safety profile remains vital (Rsa & Chemical, 2017). Medicinal plants with anti-inflammatory activity are considerably employed in the traditional treatment of several disorders of inflammation. The inflammatory response involves a complex of enzyme activation, mediator release, fluid extravasations, cell migration, tissue breakdown and repair which are aimed at host defense and usually activated in most disease conditions. These different reactions in the inflammatory response cascade are therapeutic targets which anti-inflammatory agents including medicinal plants interfere with to suppress exacerbated inflammatory responses usually invoked in such disorders as rheumatoid arthritis in infection or injury (Osemwegie & Dahunsi, 2015).

anti-inflammatory drugs are often not suitable in all cases, particularly chronic pain due to its side effects and tolerance (Traore *et al.*, 2013). Medicinal plants are known to be an important source of new chemical substances with potential therapeutic effects (Abioye *et al.*, 2013). The increase in the indiscriminate use of plant extracts is further aggravated by the belief that plants are relatively safe for consumption simply because they are of natural origin. However, the consumption of medicinal plants as conventional plants or as curatives may cause adverse toxicological effects to human health (Jordan *et al.*, 2010). In recent years, the pharmacological and toxicological effects of these plants have begun to receive attention from scientist for the verification of their acclaimed pharmacological and therapeutic properties (Rebecca, 2002). A proper scientific evaluation of medicinal plants with pertinent emphasis on toxicological paradigms is imperative while assessing the efficacy (Karou *et al.*, 2011). The current conventional therapy for pain relief poses some side effects to humans hence there is an urgent need for safe, affordable and effective analgesic and anti-inflammatory drugs (Adetutu, *et al.*, 2011).

The plants are found to grow in hot tropical climates where they are found at elevations from around 200 metres above sea level (Komolafe *et al.*, 2017). *S. latifolius* is a deciduous Tree growing to 10 m (32ft) by 10 m (32ft) at a medium rate. They grow best in areas where the mean annual temperature is around 27°C and a range of moisture conditions from fairly dry savannah to moist forest preferring a mean annual rainfall around 2,700mm (Berde & Sethna, 2002). The plants are tolerant of a range of soils, preferring a position in full sun which makes them very drought tolerant.

## 2. MATERIALS AND METHODS

### Sample collection and identification

The roots of *Sarcocephalus latifolius* was obtained from Senchi village, Zuru Local Government kebbi state Nigeria. The sample was then taken to the department of plant and biotechnology kebbi State University of Science and Technology Aliero for identification.

### Preparation of plant sample

The roots of *Sarcocephalus latifolius* was washed with clean water to remove impurities and air dried under room temperature after which it was be pulverized into powdered form using mortar and pestle. The powdered sample was then weighed with a weighing balance and kept in a clean specimen container for safe keeping before use.

### Preparation of extract

The methanol extract of *Sarcocephalus latifolius* was prepared by soaking 200g of the powdered sample with 500mls methanol and allowed for 72 hours. There after the extract obtained was filtered using a filter paper of porous size and the filtrate concentrated using rotary evaporator at 45°C.

### Experimental animals

Adult albino rats of both sexes were used for the experiment. Rats weighing between 150-200g/w were purchased from NARICT Zaria Kaduna State. All animals were kept in well ventilated wooden cages under a standard laboratory condition at the animal house of the Kebbi state university of science and technology Aliero Kebbi state Nigeria. The rats were allowed free access to food and water. They were also kept under good laboratory condition for about one week to acclimatize with the laboratory environment before commencement of the research. The research was conducted in compliance with the WHO guidelines and internationally accepted principles of laboratory animal use. Animal care guidelines on animal use protocol review of Canadian council on animal care (CCAC, 1997 and OECD 2001).

### Anti-inflammatory activity

#### Carrageen induced paw edema

Oedema was induced by injecting 0.1ml of 1% w/v carragenan suspension into the sub-planter region of the right hind paw of the rats according to the method described by winter *et al.*, (1962). The control group A received 0.5ml normal saline while the control group B received 10mg/kg diclofenac. The test groups received 100, 200 and 300 mg/kg of *sarcocephalus latifolius* extract respectively 1 hour before carragenan was administered. Measurement of the paw size was carried out by wrapping a piece of cotton thread round the paw and the length of the thread corresponding to the paw circumference was determined using meter rule (Hess and Milonig, 1972; Bamgbose and Naomesi, 1981). The inhibitory activity was calculated according to the following formular.

$$\% \text{ inhibition} = \frac{(C_1 - C_0)_{\text{control}} - (C_1 - C_0)_{\text{treated}}}{(C_1 - C_0)_{\text{control}}} \times 100$$

Where  $C_1$  is the paw circumference at time  $t$ ,  $C_0$  is the paw circumference before carrageen injection,  $(C_1 - C_0)$  is oedema,  $(C_1 - C_0)$  control is the oedema or paw size after carrageen injection to control rats at time  $t$ . In practice, carrageen activity is maximum at 3 hours and the effect of the extract at that time is accepted as the optimum inhibitory effect.

### Toxicity studies

#### Acute oral toxicity assay of *Sarcocephalus latifolius*

The acute oral toxicity of methanol root extract of *S. latifolius* was conducted using wister albino rats as described by Lorke (1983). The studies was conducted in two stages,

PHASE I: in the first phase of the studies nine (9) rats were grouped into three each having three (3) rats. The rats were weighed and their doses were computed with respect to their body weight. The prepared crude extract was then administered with 10, 100 and 1000 mg/kg body weight respectively. After that the rats were observed for about 30 minutes for any sign of toxicity or mortality and further observation made for 3 to 4 hours, then 8 hours and for 24 hours. Subsequently for 72 hours and thereafter daily for 14 days after administration of the extract. Zero record of mortality will lead to phase II of the assay.

Group	Number of rats	Dose (mg/kg)
I	Three (3)	10
II	Three (3)	100
III	Three (3)	1000

Phase II: in phase II, 4 rats were separated into 4 groups I-IV the rats received 1200, 1600, 2900 and 5000 mg/kg body weight of the methanol extract of *S. latifolius* orally after which they were observed for 30 minutes for signs of toxicity or mortality. Further observation was made for 3 to 4 hours, 72 hours and subsequently for 14 days after administration.

Group	Number of rats	Dose (mg/kg)
I	One (1)	1200
II	One (1)	1600
III	One (1)	2900
IV	One (1)	5000

## 3. RESULTS AND DISCUSSION

### RESULTS

#### Anti-inflammatory activity of Methanol root extract of *S. Latifolius*

Dose Administration (mg/kg)	Anti-inflammatory activity (%)
Normal saline (0.5ml/kg)	0.83±0.50 <sup>a</sup>
Diclofenac (10mg/kg)	0.54±0.97 <sup>d</sup>
<i>S. Latifolius</i> (100mg/kg)	0.73±0.86 <sup>b</sup>
<i>S. Latifolius</i> (200mg/kg)	0.70±0.40 <sup>bc</sup>
<i>S. Latifolius</i> (300mg/kg)	0.41±0.51 <sup>c</sup>

Values are expressed as mean ± standard error of mean,  $n = 5$ , mean values having common superscript letters in a column are not significantly different ( $P < .05$ ) analyzed with SPSS Version 20.0 (one-way ANOVA followed by Duncan's multiple range test).

## DISCUSSION

Natural product remedies are popular and are gaining a lot of acceptance among people in the prevention of diseases and ailments owing to their proximity and cost effectiveness. The general belief that products of natural origin are relatively safe and free from side effects which synthetic drugs poses drives people towards the use of this natural products for medicine (Raghunath, 2014). However, many of the natural medicines in the market do not have sufficient scientific safety data and toxicological certification. This is therefore important since all natural medicine is taken under self medication and administration without medical advice or supervision. Hence the proper knowledge on the toxicity and safety profile of the natural medicine has become crucial and necessary.

Recent research on the consumption of medicinal plants as conventional medicines or as curative revealed that excessive consumption causes adverse toxicological effects to human health (Quaile *et al.*, 2010). This research showed no sign of toxicity or mortality at dose 10, 100 and 1000mg/kg in rats during the acute toxicity assay. However the animals were observed to be dull for 3-4 hours after administration of the extract then later they were seen to resume their normal activities. This suggests that the LD<sub>50</sub> of the plant extract is greater than or equal to 5000mg/kg. Similarly in the sub-chronic toxicity assay, there was mortality at different doses of the experiment.

According to (Ogbe *et al.*, 2010), reduction in body weight is considered a sensitive indication of toxicity after exposure to a toxic substance. There were signs of toxicity observed and deaths recorded among the experimental animals during the 28 days of daily administration of the plant extract. Among was general body weakness in all the treated groups coupled with loss of appetite when compared to the control. Much signs of toxicity was observed among group IV and V that took higher dose at 750mg/kg and 1000mg/kg. After 7 days of administration, there was massive loss in weight of the animals as a result of loss of appetite due to the toxic nature of the plant extract that affected their eating habit. The weight of the animals reduced greatly when compared to that of the control group. This suggests that the plant extract is highly toxic when consumed for a long period of time.

Further assessment of the toxicity profile was seen in the hematological parameters and pathological status of the experimental animals and humans as it can be a pointer to the direct effect caused by the extract (Adeneye *et al.*, 2006). In herbal toxicity studies, elevation in the level of WBC, LYM and NEUT may be an indicator that the plant extract have induced the immune response of the treated animals (Tousson *et al.*, 2011). On the other hand a significant ( $P < 0.05$ ) decrease in these parameters of the blood may indicate that there is no sufficient production of leucocytes which means the body is susceptible to disease and infection and is less likely to fight infections. Thus the hematological analysis of the study revealed a significant ( $P < 0.05$ ) decrease on the level of WBC, RBC and LYMP at 100 and 200 mg/kg when compared to the control. These results suggest that the plant extract of *S. Latifolius* possesses a chemical constituent capable of diminishing the production of leucocytes or suppresses its activity (WEINGAND *et al.*, 1996).

According to (Peters *et al.*, 2008), packed cell volume (PCV) or Hematocrits (HCT), haemoglobin (HGB), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), are major indices for evaluating circulatory erythrocytes and are significant in the diagnosis of anemia and serve as a useful pointer for bone marrow capacity to produce red blood cells in mammals (Chineke, Ologun, & Ikeobi, 2006). The effect of *S. Latifolius* showed a significant ( $P < 0.05$ ) increase in MCH and MCHC while MCV, MPV HGB and PCT reduced significantly ( $P < 0.05$ ) in the treated rats compared to the control group. These results revealed that the plant *S. Latifolius* may have caused a remarkable toxic effect on the red blood cells and blood parameters analyzed at the respective doses. It also suggests that since the immune producers are suppressed, animals may be at risk of developing an ailment such as anemia.

The primary organs prone to toxic effects are the kidney and liver both of which play an important role in assessing the changes in biochemical parameters. A mild inflammation or tissue damage to these organs is an indication that the permeability of cell membrane will increase and cytoplasmic enzymes such as LD, ALD and AST will be released while necrosis will release mitochondrial ALT as well as AST leaking into the blood and increase in levels (Hall *et al.*, 2012). The biochemical indices monitored in serum such as the electrolytes and other secretory substances of the liver and the kidney can be used as markers for assessing the functional capacities of the organs (Nafiu, Akanji, & Yakubu, 2011). These parameters if altered will affect or impair the normal functioning of the organs. In this present study, there is a significant ( $P < 0.05$ ) increase in ALP, AST and ALP at dose 100 and 200 mg/kg which indicates liver injury from damage or inflamed liver. Although it is said that in clinical practice, it is not out of place to see an elevated level of ALT and AST in non hepatic conditions such as myocardial infarction. The significant ( $P < 0.05$ ) increase may be due to heart attack

or damaged hepatic cells and it may have been induced by some phyto compounds in the plant extract (Papafragkakis *et al.*, 2014). ALP is a marker enzyme for the plasma membrane and endoplasmic reticulum. The ( $P < 0.05$ ) significant increase in ALP could be due to renal damage and inflammation. The elevation of ALP in this research can be attributed to the presence of some phyto compounds in the plant extract of *S. Latifolius*.

The extract was found to inhibit carrageen induced paw oedema significantly ( $P < 0.05$ ) by inhibiting mediators of inflammation (Shivalingaiah, 2016). Although carrageen induced paw oedema is a test of acute inflammation, the result obtained showed that the roots extract of *S. Latifolius* can effectively reduce inflammation. This suggests that the extract possess both analgesic and anti-inflammatory activity that might be mediated through a common mechanism. Analgesic and Anti-inflammatory activities are commonly possessed by non-steroidal anti-inflammatory drugs (NSAIDs). These NSAIDs exert their effects principally by inhibiting the synthesis of prostaglandin (Vane, 1971), an eicosanoid mediator of the inflammatory response (Yun *et al.*, 2008). In addition to their inflammatory response, prostaglandin also causes pain and sensitizes the skin to painful stimuli (Dray, 1995).

#### 4. CONCLUSION

This present research showed that methanol root extract of *Sarcocephalus Latifolius* has anti-inflammatory activity by significantly reduce carrageen induced paw oedema in rats. It can then be concluded that methanol root extract of *Sarcocephalus Latifolius* can be used in traditional medicine in the treatment of inflammation and related diseases haven been validated in this research and thus it is recommended for use as formulations in traditional medicine.

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