

RESEARCH ARTICLE

ACUTE TOXICITY (LD₅₀) STUDY OF METHANOL EXTRACT OF SALACIA SENEGALENSIS Lam (DC) LEAF ON ALBINO MICE

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Abstract

Salacia senegalensis is an acclaimed medicinal plant use locally by the people of the South-East zone Nigeria in the treatment of malaria, skin problem and lotion for sick children. Therefore, acute toxicity study (LD₅₀) was carried out on methanol extract of its leaf. Results of phase I and phase II of the study showed no mortality was recorded in any of the experimental groups of mice in 24 hours (a day), 72 hours (3days) and up to four weeks after oral and intra-peritoneal administration of 5000mg per kg body weight of extract. Also the histology (photomicrograph) of the liver sections showed no histopathological lesion, showed multinucleation, a large but centrally positioned hepatic vein, pleomorphic nuclei, normal sinusoids with intact hepatic cyto-architecture are being regenerated (H & E X 400). Hence, oral and intra-peritoneal administration of the extract at a dose of less than or equal to 5000mg per kg body weight is safe, but extremely high doses may not be advisable.

Keywords: Acute Toxicity, oral and intraperitoneal administration, *salacia senegalensis*.

Introduction

Salacia Senegalensis Lam (DC) is a shrub erect or climbing with white or pale greenish cream petals and orange or yellow flowers. It is found in forest re-growth. It belongs to the family *Celastraceae*.

Medicinally, the plant (leaves and fruits are mainly used) is mainly used in treatment of skin problem and malaria (anti-malarial) (NNMDA, 2011). Also, infusion of the leaves could serve as lotion for sick children (NNMDA, 2011). Lethal toxicity (acute toxicity) is the ability of a chemical to cause ill effect "relatively soon" after one oral administration or a 4 – hour exposure of a chemical in air (Senin, 2006). "Relatively soon," is usually defined as a period of minutes, hours (24) or days (up to about 2 weeks) but rarely longer (Senin, 2006). LD stands for "Lethal dose." LD₅₀ is the amount of material, given all at once, which causes the death of 50% of a group of test animals. The LD₅₀ is one way to

measure the short – term poisoning potential (acute toxicity) (Senin, 2006). Toxicologists can use many kinds of animals but most often testing is done with rats and mice. It is usually expressed as the amount of chemical administered (e.g. milligrams) per 100grams (for smaller animals) or per kilogram (for bigger subjects) of the body weight of the test animal (Gadanya, *et al.*, 2011). The LD₅₀ can be found for any route of entry or administration, but dermal (intra-peritoneal) and oral administration methods are the most common. The LD₅₀ value obtained at the end of the experiment is identified as LD₅₀ (oral), LD₅₀ (skin) etc. as appropriate. The most frequently performed lethality study is the oral LD₅₀. The results of oral studies are important for drugs, food and accidental domestic poisonings. In general, the smaller the LD₅₀ value, the more toxic the chemical is. Also, the larger the LD₅₀ value, the lower the toxicity (Senin, 2006). LD₅₀ value can be

compared to other values using a toxicity scale. Confusion sometimes occurs because several different toxicity scales are in use. The two most common scales used are the "Hodge and Sterner scale" and the "Gosselin, Smith and Hodge scale (Senin, 2006)." These tables/scales differ in both the numerical rating given to each class and the terms used to describe each class. It is important to know that the actual LD₅₀ value may be different for a given chemical depending on the route of exposure (Oral, dermal, inhalation) (Senin, 2006).

Therefore, this research work is aimed at finding out the oral and intra-peritoneal LD₅₀ value of the *salacia senegalensis*- a medicinal plant of south-east zone Nigeria.

Materials and Methods

Sample collection

The medicinal plant *salacia senegalensis* was obtained from the re-grown forest at Orji Owerri North L.G.A, Imo State, Nigeria, identified and authenticated by renowned taxonomists of the department of Plant Science and Biotechnology Imo State University, Owerri, Nigeria.

Sample preparation and extraction

The leaves of *Salacia senegalensis* were dried at room temperature (25°C) and milled into powder using clean and sterile milling machine. The milled sample was soaked in 95% methanol for 72hours (3days). After which it was filtered using Whatman no. 48 filter paper. The filtrates were concentrated under rotary vacuum evaporator (RE 2000) and were further dried in water bath at 60°C at the Malaria research centre University of Port-Harcourt Project B, Rivers State, Nigeria.

Determination of Acute toxicity (LD₅₀)

The method of Lorke (1983) was used in LD₅₀ determination. Three groups of four mice each were orally administered methanol leaf extract of *Salacia senegalensis* weighing 10mg, 100mg and 1000mg per kg body weight and were observed for 24hours, 72hours, two weeks and four weeks as shown in the **tables 1**. In the second phase, four groups of one mouse each were administered orally with the extract at doses of 1600mg, 2900mg, and 5000mg per kg body weight as shown in the **tables 3**. The mice were observed for 24hours, 72hours, two weeks and four weeks and number of death

was recorded and LD₅₀ value was determined. The same procedure was repeated for intra-peritoneal administration in mice as shown in **tables 2** and **4** below.

Histology Study

The mice were sacrificed and their livers obtained and stored with 40% formalin. This was later processed and embedded into paraffin wax. The tissue block sectioned into 5µm, was later stained with heamatoxylin and eosine and their photomicrograph taken (as shown in the figure 1 below).

Results and Discussion

From the result of acute toxicity study of *Salacia senegalensis* (Tables 1 to 4), no mortality was recorded in any of the experimental groups in 24hours, 72hours and up to four weeks after oral and intra-peritoneal administration of 5000mg per kg body weight of the extract. According to toxicity classes of Hodge and Sterner (2005), and Lorke (1983) any compound with oral LD₅₀ (rat) of 5000mg/kg or more should be considered as practically harmless. A preliminary study has shown that this plant contains alkaloids, and saponins (unpublished yet). Saponins enhance nutrient absorption and aid in animal digestion. Significant toxicity is usually as a result of suicide attempt or inappropriate self-administration for therapeutic purposes (Raffi and Mark, 2009). Also, alkaloids have some pharmacological effects and are used as medications, recreational drugs, or in entheogenic rituals e.g. the local anesthetic and stimulant cocaine, the stimulant caffeine, the analgesic morphine or the antimalarial drug quinine (Tailang and Sharma, 2009). Also the histology (photomicrograph) of the liver sections showed no histopathological lesion, showed multinucleation, a large but centrally positioned hepatic vein, pleomorphic nuclei, normal sinusoids with intact hepatic cyto-architecture are being regenerated (H & E X 400) as shown in figure 1. Hence, intra-peritoneal or oral administration of *Salacia senegalensis* extract at a dose of less than or equal to 5000mg per kg body weight is safe, but extremely high doses may not be advisable.

Conclusion

The acute toxicity study showed that administration of *Salacia senegalensis* extract at a dose of less than or equal to 5000mg per kg body weight is safe, but extreme high doses may not be advisable.

Table 1. Mortality recorded in oral Lethal Dose (LD₅₀) determination for the extract

Dose(mg/kg body weight)	Methanol extract of <i>Salacia senegalensis</i>
10	$\frac{0}{4}$
100	$\frac{0}{4}$
1000	$\frac{0}{4}$
Control	$\frac{0}{4}$

$(\frac{0}{4})$ 0 = number of death, 4 = number of mice used for the test

Table 2. Mortality recorded in intra-peritoneal Lethal Dose (LD₅₀) determination for the extract

Dose(mg/kg body weight)	Methanol extract of <i>Salacia senegalensis</i>
10	$\frac{0}{4}$
100	$\frac{0}{4}$
1000	$\frac{0}{4}$
Control	$\frac{0}{4}$

$(\frac{0}{4})$ 0 = number of death, 4 = number of mice used for the test

Table 3. Mortality recorded in oral Lethal Dose (LD₅₀) determination for the extract

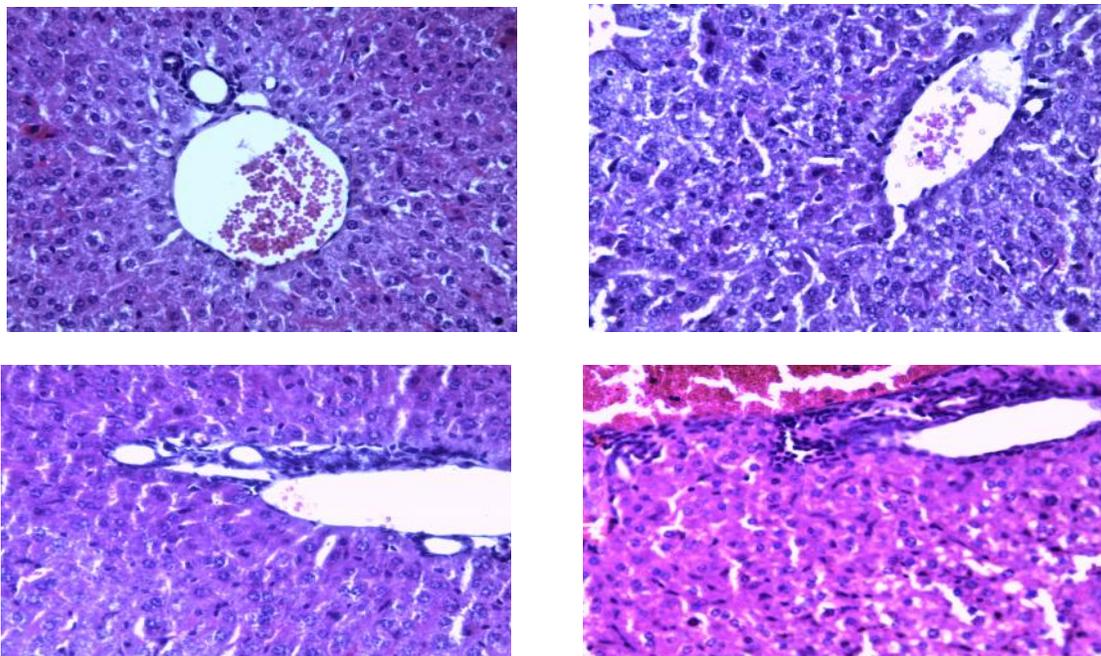
Dose(mg/kg body weight)	Methanol extract of <i>Salacia senegalensis</i>
1600	$\frac{0}{1}$
2900	$\frac{0}{1}$
5000	$\frac{0}{1}$

$(\frac{0}{1})$ 0 = number of death, 1 = number of mice used for the test

Table 4: Mortality recorded in intra-peritoneal Lethal Dose (LD₅₀) determination for the extract

Dose(mg/kg body weight)	Methanol extract of <i>Salacia senegalensis</i>
1600	$\frac{0}{1}$
2900	$\frac{0}{1}$
5000	$\frac{0}{1}$

$(\frac{0}{1})$ 0 = number of death, 1 = number of mice used for the test

Figure 1. Photomicrograph of the mice livers

References

- Gadanya A. M., Sule M. S and Atiku M. K. 2011. Acute Toxicity Study of "Gadagi" Tea on Rats *Bayero Journal of Pure and Applied Sciences*, 4(2): 147 – 149
- Hodge, A. and Sterner, B. 2005 .Toxicity classes. In:Canadian center for occupational Health and safety. Copy right @1997-2010. Retrieved from (<http://www.ccohs.ca/oshanswers/chemicals/id50.htm>) On 3/5/2010.
- Lorke, D. 1983. A new approach to tropical acute toxicity testing. *Arch. Toxicol.* 53: 275-287.
- Nigeria National Medicine Development Agency (NNMDA). 2011. *Salacia senegalensis*, Medicinal plants of South-East Zone Vol.1 pp 67.
- Raffi, k. and Mark, S. 2009.Plant poisoning, glycosides-cardiac.Continually Updated Clinical Reference. Retrieved from (<http://www.answers.com/topic/glycosides>)on 21/3/2010.
- Senin, R. 2006. Acute toxicity study. Retrieved from (<http://www.ccohs.ca/oshanswers/chemicals/ld50.html>) on 27/3/2010.
- Tailang, M. and Sharma, A.K. 2009. Phytochemistry (theory and practicals). Birla publications, India.Pp.229-237.