Evaluation of the Anti-snake Venom Activity of Leaf Extract of *Sansevieria liberica* ger. & labr (Agavaceae.) in Mice

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Abstract: The prevalence of snake bite and death from snake bite envenomation is becoming a serious public health problem globally. The cost, non availability and the adverse reactions associated with antisnake venom serum has encouraged the use of medicinal herbs by traditional healers in the treatment of snakebites. In this study we evaluated the antisnake venom activity of the leaves of *Sansevieria liberica* against *Naja naja nigricollis* venom in mice. *S. liberica* is very popular among the traditional healers in south east Nigeria for the treatment of snake bites. The ground fresh leaves were extracted with ethanol and a portion of the ethanol extract was fractionated with n-hexane, ethyl acetate and butanol to afford the respective solvent fractions. The extract was subjected to acute toxicity (LD₅₀) and phytochemical testing. The LD₅₀ of the venom was similarly determined. The antisnake venom activity of the extract/fraction was determined against the LD₅₀ (353.5 ug/kg) and double the LD₅₀ (707 ug/kg) of the venom. The effect of the extract on bleeding and clotting time of the venom-intoxicated mice was investigated. Also studied was the effect of the extract on acetylcholine-induced contraction of the isolated frog rectus abdominus. The extract/fractions significantly protected the mice from *Naja naja nigricollis* venom-induced mortality in mice. The bleeding and clotting time of the venom-intoxicated rats were significantly (p < 0.05) decreased by the extract/fractions. The acetylcholine-induced contraction of frog rectus abdominus was significantly inhibited by the extract. These results suggest that the leaves of *S. liberica* exhibit antisnake venom properties that could be harnessed in treating patients with snakebite envenomation.

Keywords: *Sansevieria liberica*, *Naja naja nigricollis*, Bleeding Time, Clotting Time, Frog Rectus Abdominus

Introduction

Envenomation resulting from snake bites is an important public health problem in rural areas of tropical and sub-tropical countries of Africa, Asia, Oceania and Latin America (Chippaux 1998). It is a major socio-medical problem of south east Asia and sub-sahara African countries. About 216 species of snake occur in India alone, and about 25 % of which is poisonous (Bawaskar, 2004). Poisonous snake species like *Echis carinatus*, *Naja naja*, *Daboia russelli*, *Bungarus caeruleus* and *Ophiophagus hannah* account for the majority of the bites and mortality (Gomes et al, 2010). The exact epidemiology as well as number of deaths from snake bites is difficult to obtain. This is partly due to the fact that most of the snake bites occur in the rural areas, and also the dependence on traditional healers.

Snake venom is a very complex poison comprising a mixture of enzymatic and non-enzymatic toxic compounds and other non-toxic proteins, carbohydrates and metals. The poison components can incorporate proteases, nuclease, phosphodiesterases and other substances which alter cell functions and physiological processes (Sajon et al, 2017). The venom toxins are mainly neurotoxins, cytotoxins, myotoxins and cardiotoxins which evoke the variety of adverse reactions and death associated with snake venoms.

The treatment of snake bite varies as the bite and snake species. Antivenom immunotherapy remains the specific treatment against snake bite. In most poor resource countries where incidentally, snake bites are frequent, antivenoms are expensive and limited in supply. The mode of administration, usually parenterally, is another limitation. Most antivenoms are not broad spectrum, and their effectiveness may depend on the type of snake involved and the time lag before administration. Antivenom therapy is associated with a number of side effects like anaphylactic shock, pyrogen reaction and serum sickness (Maya Devis et al, 2002).

The plant kingdom offers alternative option for the management of snake bites. Over the years many attempts have been made for the development of...
snake venom antidotes from plants sources. In most rural areas, traditional healers are the first port of call in cases of snake bite, and some of these cases were successfully treated with folk medicines, especially medicinal plants. Several medicinal plants, which appear in old drug recipes or which have been passed on by oral tradition, are believed to be snakebite antidotes and are recommended for the treatment of snake bite (Alam and Gomes, 2003; dos Santos Gomes et al, 2010; Kaushik et al, 2013). A number of medicinal plants including Jatropha species (Gomes et al, 2016; Felix-Silva et al, 2018), Asystasia gangetica (Enenebeaku et al, 2018), Parkia biglobosa (Asuzu and Harvey, 2003), Alyosia citriodora (Caceres et al, 2017), Sapindas saponaria (da Silva et al, 2012), Albizia lebbeck (Amog et al, 2016), Carissa spinarum (Janardhan et al, 2015), Piper longum (Shenoy et al, 2013) and Crinum jagus (Zadani et al, 2018) have been reported to possess antivenom properties.

In the eastern part of Nigeria, the leaves of Sansevieria liberica ger.& labr is a common folk remedy for the treatment of snake bite, irrespective of the species of snake involved. Traditional healers in the area have attested to the usefulness and effectiveness of S. liberica leaves in the treatment of snake bite.

**Sansevieria liberica** belongs to the family Agavaceae. It is one of the bowstring hemp species and a pretty plant with bright green leaves growing 45 to 100 cm tall, with a smooth texture and light grey tip (Evans, 2005). The leaves are typically arranged in a rosette (Chahinian, 2005). It is known as “Moda” (Hausa), “Ebubagu” (Igbo) and “Ijo-ikoko” in (Yoruba) tribes of Nigeria. The common English name is Bow string hemp. The medicinal uses of S. liberica depend on the region. In the Northern part of Nigeria, it is used in traditional medicine to treat menorrhagia and menstrual pains, and to normalise abnormal menstrual period (Sambo and Ali, 2008). In the Western and Eastern parts of Nigeria it is used to treat diarrhoea, abdominal pains, gonorrhoea, eczema, pile, snake bite, impotence, asthma, and high blood pressure (Odugbemi, 2008).

The anti-inflammatory (Ratheesh, 2007; Eze et al, 2011), anti-anaemic (Ikwuchu et al, 2010), sedative and anti-convulsant activities (Adeyemi et al, 2007) of the leaves and roots have been reported. Also reported are the antidiarrhoeal (Adedeyemi, et al, 2009), antihypertensive (Ikwuchu et al, 2011), analgesic (Umukoro et al, 2008), in vitro antitrypanosomal, antileishmanial and antiplasmodial (Bero et al, 2009, 2011), diuretic (Omodamo and Jimo, 2017) and anti-oxidant (Ikwuchu et al, 2013) activities.

To the best of our knowledge, there is no scientific report on the anti-snake venom potentials of S. liberica. The aim of this study was to evaluate the anti-snake venom activity of the leaves of the plant against *Naja n.nigricolli* venom in rodents.

**Materials And Methods**

**Collection and authentication of plant materials**

Fresh leaves of *S. liberica* were harvested from Ekwulobia in Aguata Local Government Area of Anambra State Nigeria and were identified Mr. Patrick Ugwuozo of the Department of Botany, Nnamdi Azikiwe University, Awka Anambra State, Nigeria. Voucher specimen No. NAU 456 was deposited at the Herbarium of the Department of Pharmacognosy, Nnamdi Azikiwe University, Awka, Nigeria.

**Preparation of plant materials**

The methods used traditionally to prepare the plant materials which include powdering of the plant material followed by solvent (aqueous or organic) extraction was adopted. The fresh leaves were washed clean, cut into smaller pieces and pounded with a mortar and pestle. About 400 g of ground wet leaves was macerated with 2.5 litres of ethanol for 48 hours. The resulting solution was filtered using muslin cloth and concentrated under vacuum using rotary evaporator. The extract was further concentrated to dryness using water bath at 50 °C to yield 24.53g (6.13%) of crude extract. Using solid-liquid method of fractionation on a silica gel the crude extract was partitioned with n-hexane, ethyl acetate and butanol to obtain n-hexane (HF), ethyl acetate (EAF) and butanol (BF) fractions respectively in order of increasing polarity.

**Snake venom**

Freeze-dried venom of *Naja. n. nigricolli* was obtained from the Department of Veterinary Medicine, Ahmadu Bello University, Zaria, Kaduna State, Nigeria.

**Animals**

Albino mice of either sex (21-25 g) were used for the study. They were obtained from the Animal House of the Department of Pharmacology and Toxicology, Nnamdi Azikiwe University, Awka, Nigeria. The animals were housed in standard laboratory cages and conditions of 12 h light/dark cycle, at room temperature. Rodent feed (Guinea Feeds Nigeria Ltd) and clean water were provided ad libitum. All animal experiments was conducted in strict compliance with NIH guide for care and use of laboratory animals (National Institute of health (NIH) (2011) Pub No: 85-23).

**Phytochemical screening**

Qualitative phytochemical analysis of the extract was carried out to identify the presence of secondary metabolites using standard methods (Harborne, 1984).
Acute toxicity studies
Acute toxicities (LD₅₀) of the extract (oral) and the snake venom (intraperitoneal) were determined using Lorke’s method (Lorke, 1983).

Anti snake venom activity
The anti-snake venom activity of the crude extract and the fractions was tested in vivo against the LD₅₀ (353.5 ug/kg) and double the LD₅₀ (707 ug/kg) of the snake venom. The extract and fractions were administered at the dose of 125, 250 and 500 mg/kg orally. For this study, a total of 100 mice were used. The mice were divided into 4 groups (1-4). Group 1 served as the control, and received distilled water plus the two doses of the venom respectively. Groups 2-4 were used for immediate (concomitant), prophylaxis and curative treatments respectively. They were each subdivided into 6 (n=5) and treated as follows:

Group 1 (control)
1A: Distilled water (5 ml/kg) + venom (353.5 ug/kg).
1B: Distilled water (5 ml/kg) + venom (707 ug/kg).

Group 2. The venom was administered concomitantly with the extract.
2 A: Extract (125 mg/kg) + venom (353.5 ug/kg).
2 B: Extract (250 mg/kg) + venom (353.5 ug/kg).
2C: Extract (500 mg/kg) + venom (353.5 ug/kg).
2D: Extract (125 mg/kg) + venom (707 ug/kg).
2E: Extract (250 mg/kg) + venom (707 ug/kg).
2F: Extract (500 mg/kg) + venom (707 ug/kg).

Group 3. The venom was administered 1 hour after the extract (Prophylaxis)
3A: Extract (125 mg/kg) + venom (353.5 ug/kg).
3B: Extract (250 mg/kg) + venom (353.5 ug/kg).
3C: Extract (500 mg/kg) + venom (353.5 ug/kg).
3D: Extract (125 mg/kg) + venom (707 ug/kg).
3E: Extract (250 mg/kg) + venom (707 ug/kg).
3F: Extract (500 mg/kg) + venom (707 ug/kg).

Group 4. The extract was administered 30 minutes after the venom.
4A: Venom (353.5 ug/kg) + extract (125 mg/kg).
4B: Venom (353.5 ug/kg) + extract (250 mg/kg).
4C: Venom (353.5 ug/kg) + extract (500 mg/kg).
4D: Venom (707 ug/kg) + extract (125 mg/kg).
4E: Venom (707 ug/kg) + extract (250 mg/kg).
4F: Venom (707 ug/kg) + extract (500 mg/kg).

Anti snake venom activity of the fractions
The fractions (125 mg/kg p o) and the venom (707 ug/kg ip) were administered immediately (concurrently). The animals were observed for mortality for 24 hours.

Evaluation of bleeding
Twenty five (25) adult albino mice grouped into five (n=5) were used for the study. Group 1 received distilled water and severed as the control. Group 2 had the venom (707 ug/kg) alone, while groups 3-5 received the venom and the extract 125, 250 and 500 mg/kg respectively. The bleeding time was measured using the method of Mohammed et al (1969). Two hours after administering the venom (707 ug/kg), and treatment with the extract, pressure was built at the tail of the animal by stroking it with fingers. Then a sterile needle was used to puncture one of the blood vessels lying alongside of the tail. The blood that oozed out of the site was blotted gently but completely with filter paper every 15 seconds until bleeding ceases. The time of the first appearance of blood and stopping of the blood flow was taken as the bleeding time.

Evaluation of clotting time
This study involved twenty five (25) adult albino mice grouped into five (n=5). Group 1 received distilled water and severed as the control. Group 2 had the venom (707 ug/kg) alone, while groups 3-5 received the venom and the extract 125, 250 and 500 mg/kg respectively.

The clotting time was evaluated using capillary coagulation method. Non-heparinized capillary tube was used to collect blood from the retro-orbital plexus of the animal through capillary action until whole length of the tube was filled with blood. At 15 seconds interval, one half of the capillary tube was carefully broken off at one end and gently pulled apart to look for the fill in thread which is an indication of clotting. The breaking of the tube was done at 15 sec interval until fibrin thread was seen. The time it took the thread to form is taken as the clotting time (Igboechi and Anuforo, 1986).

In vitro study
Effect on frog rectus abdominus
Adult frog were killed and cut open. The rectus abdominus was obtained and mounted in an organ bath containing Frog Ringer solution of the following composition gram/litre NaCl (6.0), KCl (0.075), CaCl₂ (0.100), NaHCO₃ (0.100) and aerated with air and maintained at room temperature. The resting tension on the tissue was 1.5 g. The tissue was equilibrated for 30 minutes before graded responses to acetylcholine were obtained. Responses to the sub-maximum concentration of Ach were re-established in the pressure of 10 and 20 mg of the extract after 15 minutes incubation. The responses were recorded on Unirecorder 7050 (Ugo Basile, Italy) through an isotonic Transducer, 7004 (Ugo Basile, Italy).

Statistical analysis
Results were presented as mean ± Standard error of mean (SEM). Data were subjected to one way analyses of variance (ANOVA) followed by Post-hoc Turkey’s test for multiple comparison. Statistical package for social science (SPSS-20) was used for data analyses. P < 0.05 was considered to be statistically significant.
Results

Phytochemical constituents
Qualitative phytochemical analysis revealed the presence of saponins, flavonoids and terpenoids in high amounts while alkaloids, steroids and cardiac glycosides occurred in smaller amount quantities (Table 1).

The acute toxicity test indicated that the LD50 of the extract was above 5000 mg/kg, while that of the snake venom was 353.55 μg/kg.

Effect of the extract on LD50 of the snake venom
On simultaneous administration of the extract with the venom, 100% protection was afforded by 125 and 250 mg/kg of the extract (Table 2).

In prophylactic studies, all the mice survived at the doses of 125 and 250 mg/kg of the extract. However, 80% protection was recorded at 500 mg/kg dose (Table 2).

When the extract was administered thirty minutes after exposure of animals to the LD50 dose of the snake venom, all the animals treated with 125 mg/kg of the extract were protected while 80% protection was recorded at 250 and 500 mg/kg doses (Table 2).

Effect of the extract on double LD50 (707 μg/kg) of the snake venom:
The effect of extract on the double LD50 of the venom is shown in Table 3. The extract was more effective prophylactically as all the mice survived at all the 3 doses of the extract tested, when the extract was administered thirty minutes after exposure of the animals to twice the LD50 of the snake venom, the percent protection was dose dependent (Table 3).

Table 1: Phytochemistry of ethanol leaves extract of Sansiveria liberica

| Phytochemical constituents | Amount  |
|---------------------------|---------|
| Alkaloids                 | +       |
| Saponins                  | ++      |
| Tannins                   | -       |
| Flavonoids                | ++      |
| Steroids                  | +       |
| Terpenoids                | ++      |
| Cardiac Glycosides        | +       |
| Proteins                  | -       |
| Carbohydrates             | -       |

(+) present in small concentration, (+++) present in high concentration, (-) absent

Acute toxicity test

Table 2. Effect of the extract on the LD50 of the snake venom

| Treatment                        | Dose (mg/kg) | No of animals | Mortality | % survival |
|----------------------------------|--------------|---------------|-----------|------------|
| Control (distilled water)        | 5ml/kg       | 5             | 3         | 40         |
| Concomitant Extract              | 125          | 5             | 0         | 100        |
|                                  | 250          | 5             | 0         | 100        |
|                                  | 500          | 5             | 1         | 80         |
| Prophylaxis                      |              |               |           |            |
| Extract                          | 125          | 5             | 0         | 100        |
|                                  | 250          | 5             | 0         | 100        |
|                                  | 500          | 5             | 0         | 100        |
| 30 minutes after administration of snake venom |              |               |           |            |
| Extract                          | 125          | 5             | 0         | 100        |
|                                  | 250          | 5             | 1         | 80         |
|                                  | 500          | 5             | 1         | 80         |

Effect of the extract on double LD50 (707 μg/kg) of the snake venom:

When the extract was administered thirty minutes after exposure of the animals to twice the LD50 of the snake venom, the percent protection was dose dependent (Table 3).
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Table 3. Effect of the extracts on double LD_{50} (707 ug/kg) of the snake venom

| Treatment                  | Doses (mg/kg) | No of animals | No of Death | Survival | % survival |
|----------------------------|---------------|---------------|-------------|----------|------------|
| Control (distilled water)  | 5ml/kg        | 5             | 5           | 0        | 0          |
| Concomitant Extract        | 125           | 5             | 0           | 5        | 60         |
|                            | 250           | 5             | 1           | 4        | 80         |
|                            | 500           | 5             | 1           | 4        | 80         |
| Prophylaxis Extract        | 125           | 5             | 0           | 5        | 100        |
|                            | 250           | 5             | 0           | 5        | 100        |
|                            | 500           | 5             | 0           | 5        | 100        |

30 minutes after administration of snake venom

| Extract                  | Doses mg/kg | No of animals | No of Death | Survival | % Protection |
|--------------------------|-------------|---------------|-------------|----------|--------------|
|                          | 125         | 5             | 3           | 2        | 40           |
|                          | 250         | 5             | 2           | 3        | 60           |
|                          | 500         | 5             | 0           | 5        | 100          |

Anti-snake venom effect of the fraction
The ethyl acetate fraction (125 mg/kg) protected 100% of the animals from the 707 ug/kg dose of the venom while the n-hexane and butanol fractions at the same dose afforded 60 and 40 % protection respectively. (Table 4).

Table 4. Effect of the fractions on snake venom

| Treatments                  | Dose mg/kg | No of animals | No of death | Survival | % Protection |
|-----------------------------|------------|---------------|-------------|----------|--------------|
| Control (distilled water)   | 5 ml/kg    | 5             | 5/5         | 0/5      | 0            |
| n-hexane fraction           | 125        | 5             | 2/5         | 3/5      | 60           |
| Ethyl acetate fraction      | 125        | 5             | 0/5         | 5/5      | 100          |
| Butanol fraction            | 125        | 5             | 3/5         | 2/5      | 40           |

Effect of the extract on bleeding time.
There was a significant (p <0.05) and dose-dependent reduction in the venom-induced haemorrhage in mice. (Table 5).

Table 5. Effect of the extract on bleeding time

| Treatment                  | Bleeding time (sec) |
|----------------------------|---------------------|
| Control, (Distilled water 5 ml/kg) | 1.1 ± 0.14          |
| Snake venom (707 ug/kg) alone   | 2.2 ± 0.08          |
| Extract 125 mg/kg             | 1.0 ± 0.14*         |
| Extract 250 mg/kg             | 0.8 ± 0.27*         |
| Extract 500 mg/kg             | 0.5 ± 0.06*         |

Values are presented as mean ± Standard error of mean (SEM), n =5. *P < 0.05

Effect of the extract on clotting time
The venom-induced increase in clotting time was significantly (p < 0.05) and dose dependently decreased by the extract (Table 6).

Table 6: Effect of the wet extract on clotting time

| Treatment                  | Clotting time (sec) |
|----------------------------|---------------------|
| Control, (Distilled water 5 ml/kg) | 1.31 ± 0.14          |
| Snake venom (707 ug/kg) alone   | 2.60 ± 0.9           |
| Extract 125 mg/kg             | 1.43 ± 0.12*        |
| Extract 250 mg/kg             | 1.08 ±0.06*         |
| Extract 500 mg/kg             | 0.84 ± 0.17*        |

Values are presented as mean ± Standard error of mean (SEM), n =5. *P < 0.05:
In this study we investigated the leaves of *Sansevieria libera* for anti-snake venom activity. The results showed the ethanol extract of the leaves as a good candidate for treating snake bites. The result of the phytochemical screening of the extracts indicated the presence of some secondary metabolites that have been reported to have antivenom actions. Selvanayagam et al. (1996) reported that many phytochemical constituents like flavonoids, quinonoids, xanthene, terpenoids and polyphenol possess protein binding and enzyme inhibiting properties and also inhibit snake venom phospholipase A2 (PLA2) activity (Santosh et al., 2004).

Most snake venoms cause local and systemic bleeding which is the consequence of the damage to blood vessel walls by venom components. Therefore the ability of the extract to prevent the venom-induced hemorrhagic activity may contribute to its anti-venom activity. Similar results have been reported for *Hemidesmus indicus* (Alam et al., 1994), *Vitex negundo* and *Emblica officinalis* (Alam et al., 2003), and *Celipta prostrate* (Pithaynnkul et al., 2004).

The more potent anti-snake venom activity demonstrated by the ethylacetate fraction is indicative of the presence of more bioactive secondary metabolites in the fraction as ethylacetate has been reported by various authors to be an efficient solvent having the ability to scavenge both polar and nonpolar bioactive secondary metabolites especially alkaloids, flavonoids and saponins (Santosh et al., 2004). The ability of the extract to block acetylcholine-induced contraction of the skeletal muscle (Frog Rectus Abdominus) may be useful in ameliorating the venom-induced myotoxicity.

The results of this study indicate that the leaves of *Sansevieria libera* contain pharmacologically active compounds that could be effective in treating patients with snakebite envenomation.

**References**

1. Adeyemi O.O, Yemitan O.K, Adebisi O.O (2007). Sedative and anticonvulsant activities of the aqueous root extract *Sansevieria libera*. *Journal of Ethnopharmacology* 113 (2):111-4.
2. Adeyemi O. O, Akindele A. J, and Ogunleye E. A. (2009). “Evaluation of the antidiarrheal effect of *Sansevieria libera* root extract,” *Journal of Ethnopharmacology* 123 (3): 459–463.
3. Azu B., Abubakar M. S, Izebe K. S, Akumuka D. D and Gamaniel K. S (2005). Effect of *Anonna senegalensis* root bark extracts on *Naja nigricollis* venom in rats. *Journal of Ethnopharmacology* 96: 507-513
4. Alam M.I, Auddy, B., Gomes, A. (1994.). Isolation purification and partial characterization of Viper venom neutralizing factor from the root extract of Indian medicinal plant *Hemidesmus indicus* R. British *Toxicom.* 32: 155–1557.
5. Alam M.I and Gomes A (2003). Snake venom neutralization by Indian medicinal plants (*Vitex negundo* and *Emblica officinalis*) root extracts. *Journal of Ethnopharmacology* 86 (3): 75–80.
6. Amog P. U, Manjuprasaanna V. N, Yariswamy A. N, Nanjaraj a. N et al (2016). *Albizia lebbeck* seed metanolic extract as complimentary therapy to manage local toxicity of *Echis carinatus* venom in murine model. *Pharmaceutical Biology* 54 (11): 2568-2574.
7. Azusu I. U and Harvey A. L (2003). The antivenom activities of *Parkia biglobosa* (Mimosaceae) stem bark extract. *Toxicon* 42(7): 763-768
8. Bawaskar H.S (2004). Snake venoms and anti-venoms: critical supply issues. Journal of Association of Physicians India. 52 (2): 11–13.

9. Bero J, Ganfou J, Jonville .M.C, Michel F, Fernand .G, Patrick .D, Mansourou .M, Joelle Q.L (2009). In vitro anti-plasmodia activity of plants used in Benin in traditional medicine to treat malaria. Journal of Ethnopharmacology 122 (3): 439–444.

10. Bero J, Hannaut V, Chataigné G, Hérent M.F, Quentin-Leclercq J. (2011). In vitro antitrypanosomal and anti-leishmanial activity of plants used in Benin in traditional medicine and bio-guided fractionation of the most active extract. Journal of Ethnopharmacology 137(2): 998–1002. doi: 10.1016/j.jep.2011.07.022.

11. Boyer L.V, Seifert S.A, and Cain J.S (2001). Recurrence phenomena after immunoglobulin therapy for snake envenomations—part 2. Guidelines for clinical management with crotaline Fab antivenom. Annals of Emergency Medicine 37 (2): 196–20.

12. Caceres M. I, Ricciardi G. A, Torres A. M, Ricciardi B. V et al (2017). In vitro antitoxin venom activities of Aloysia citriodora, Palau: New possibilities for a known aromatic plant. Journal of Essential Oil Bearing Plants 20 (1): 132–140.

13. Chahinian B.J (2005). The Splendid Sansevieria: An Account of the Species. ISBN 987-43-9250-9.

14. Chatterjee I, Chakravarty A.K., and Gomes .A (2006). Dabona russellii and Naja kaouthia venom neutralization by lupeol acetate isolated from the root extract of Indian sarsaparilla Hemidesmus indicus R. Journal of Ethnopharmacology. 106: 38–43.

15. Chippaux J.P (1998). Snake Bite: Appraisal of the Global Situation. WHO Bulletin 76 (10):515-524.

16. Corrigan P, Russell F.E, and Wainschel .J (1978). “Clinical reactions to antivenom,” in Toxicon-Animal, Plant and Microbial, P. Rosenberg, Ed., 457–465, Pergamon Press, Oxford, UK.

17. Da Silva M. L, Mareussi S, Fernandes R. S, Pereira P. S et al (2012). Antisnake venom activities of extracts and fractions from callus cultures of Sapium sapomaria. Pharmaceutical Biology 50 (3): 366-375.

18. dos Santos Gomes J. A, Felix-Silva J, Fernandes M. J, Amaral J. G et al (2016). Aqueous leaf extract of Jatropha mollissima (Pohl) Bail decreases local effects induced by Bothropic venom. BioMed Research International 2016.DOI:10.1155/2016/6101742.

19. Enebeaka C. K, Umerie S. C, Nwankwo M. U and Enenebeaka C. K (2018). Antisnake venom activities of the leaf extracts of Asystasia gangetica and Newbouldia leavis (p. Beauv). Would News of Natural Sciences 16: 38-41.

20. Eze, C. C, Inya-agha, S. I, Ezugwu, C. O, Ezea, S. C. (2011). Evaluation of anti-inflammatory property of the leaves of Sansevieria liberica, Asian PacificJournal of Tropical Medicine 4(10):791–795.doi:10.1016/S1995-7645 (11)60195-8

21. Evans W.C (2005): A taxonomic approach to the study of medicinal plants and animal-derived drugs. In: Evans WC (ed.): Trease and Evans pharmacognosy, 5th ed. (15-40). India: Elsevier

22. Felix-Silva J, Souza T, Menezes Y. A. S, Cabral B et al (2014). Aqueous leaf extract of Jatropha gossypifolia L. (Euphorbiaceae) inhibits enzymatic and biological actions of Bothrops jararaca venom. PLOS ONE 9 (8) e104952.

23. Felix-Silva J, Gomes J. A. S, Fernandes J. M, Moura A. K. C et al (2018). Comparison of two Jatropha species (Euphorbiaceae) used popularly to treat snake bites in Brazil: Chemical profile, inhibitory activity against Bothrops flavoviridis venom and antibacterial activity. Journal of Ethnopharmacology 213: 12-20.

24. Gomes A, Das R, Sarkhel S, Mishra R (2010). Herbs and herbal constituents against snake bite. Indian Journal of Experimental Biology 48: 865-878.

25. Harborne J. B (1984). Phytochemical Methods: A Guide to Modern Technique of Plant Analyssys, 2nd Edn. Chapman and Hall London, P.282.

26. Harvey A (2003). Testing of natural remedies for natural toxins. Toxicon 41:939.

27. Igbecchi, A. C and Anufooro, D. C (1986). Anticoagulant activities of extracts of Epautorutor odoratum and Vernonia amygdalin. In Sofowora A (Ed), The State of Medicinal Plants Research in Nigeria. University of Ile-Ife Press, Nigeria

28. Ikewuchi CC, Ikewuchi JC, Onyeike EN and Ayalogu EO (2010). Effect of Sansevieria liberica Gérôme and Labroy on plasma chemistry and haematological indices of salt-loaded rats. Research Journal of Science and Technology. 2(5): 110-114

29. Ikewuchi C.C, Ayalogu, E. O, Onyeka, E. N, Ikewuchi J.C (2011). Effect of aqueous extract of the leaves of Sansevieria liberica on blood pressure and pulse rate of sub-chronic salt loaded rats. Journal of Natural Remedies 11(2): 30-38

30. Ikewuchi C.J (2013). Positive moderation of the hematology, plasma biochemistry and oculo indices of oxidative stress in alloxan- induced diabetic rats, by an aqueous extract of the leaves of sansevieria liberica. Asian Pacific Journal of Tropical Medicine 11 (8 ) : 467-472.

31. Janardhan B, Shikanth V. M, Mirajkar K. K, Mores S. S (2015). In vitro anti-haemorrhage activity of Carissa spinarum Linn leaf extracts. Journal of Herbs, Spices and Medicinal Plants 21 (3): 283-293.

32. Kaushik, A, Ambesajur, A, Kaushik, J. I and Girmay B (2013). Snake venom neutralization effects of African medicinal plants and their impact on snakebites: A Review. Asian Journal of Biomedical and Pharmaceutical Sciences 3(1): 1-6

33. Lorké D (1983). A new approach to practical acute toxicity testing. Archives of Toxicology 54(4): 275-287.

34. Maya Devi C, Vasanthia B.M, Vijayan L. Umashankar P.R et al (2002). An improved method for isolation of anti-viper venom antibodies from chicken egg yolk. Journal of Biochemical and Biophysics Methods. 51(4): 129–138.

35. Mohammed, A. H, Serougi, M. S, and Hanna, M. M (1969) Observations on the effects of Echis carinatus venom on blood clotting. Toxicon 6: 215-219.

36. Mors W. B (1991). Plants active against snake bite. Plants and Traditional Medicine 5: 353-373.

37. Mors, W.B. (1991). Plants active against snake bite. In: Wagner, H., Farnsworth, N.R. (Eds.), Economic and Medicinal Plant Research, 5: 353–373.

38. Morais V. M and Massaldi H (2009). Snake antivenom: Adverse reactions and production technology. Journal of Venom and Animal Toxins Including Tropical Diseases 15: 2-18.
Evaluation of the Anti-snake Venom Activity of Leaf Extract of Sansevieria liberica ger.& labr (Agavaceae.) in Mice

39. Mors, W.B., do Nascimento, M.C., Pereira, B.M.R., Pereira, N.A (2000). Plant natural products active against snake bite-the molecular approach. Phytochemistry. 55: 627–642.
40. Ode O. J and Azusu I. U (2006). The anti-snake venom activities of the mehtanolic extract of the bulb of Crinum jagus (maryllidaceae). Toxicon 48:331
41. Odagbemi ,T (2008). A Textbook of Medicinal Plants in Nigeria. 1stEdition, University of Lagos Press, Nigeria.345-395.
42. Omodamiro O, O and Jimoh M. A (2017). Evaluation of in vitro anti-oxidant, anti-inflammatory and diuretic potentials of Sansevieria liberica leaves in Wistar albino rats. The Pharmaceutical and Chemical Journal 4 (1): 16–24.
43. Pithayanukul P, Sastitorn L., Rapepol B, Narumol P, Rutt S (2004). Anti-venom potential of butanolic extract of Eclipta prostrate against Malayan pit viper venom. Journal of Ethnopharmacology, 90:347–352.
44. Ratheesh M. A (2007). Antiinflammatory activity of Sansevieria liberica on carrageenan-induced paw oedema in male rats. African Journal of Biotechnology 6 (10): 1209-1211.
45. Sajon S. R, Sana S and Rana S (2017). Anti-venoms for snake bite: A synthetic and traditional drugs review. Journal of Pharmacognosy and Phytochemistry 6 (3): 190-197
46. Sambo B.Z, Ali H.A, (2008). Some Medicinal Plants used as Traditional Recipes for some Disorders among Northern Communities in Nigeria.1st Edition, University of Lagos Press, 65.
47. Santosh R, Faltepur Shuvai, Gawade P (2004). Preliminary screening of herbal plant extracts for anti-venom activity against common seen snake (Enhydrina schistose) Poisoning Pharmacognosy Magazine 16: 56-60.
48. Selvanayagam, Z.E., Gnanavendhan, S.G., Balakrishna, K., Rao, R.B., Sivaraman, J., Subramanian, K., Puri, R., Puri, R.K. (1996). Ehretianone, a novel quinonoid xanthene from Ehretia buxifolia with antismake venom activity. Journal of Natural Products. 59: 664–667.
49. Shenoy P, A, Nipate S, S, Sonpetkat J. M, Salvi N et al (2013). Antisnake venom activities of ethanol extract of Piper longum (Piperaceae) against Russell’s viper venom: Characterization of piperine as acting principle. Journal of Ethnopharmacology 147 (2). DOI:10.1016/j.jep.2013.02.022
50. Umukoro S and Ashorobi R. B., (2008). Effects of the methanol leaf extract of Sansevieria liberica on the central nervous system in mice. Journal of Natural Remedies, 8 (2). 242–246.
51. Zadani A. H, Magaji P, K, Sarkiyayi S and Wurochekke A. U (2018). Antisnake venom activity of the aqueous and ethanolic extracts of Crinum jagus bulb. Asian Journal Research in Biochemistry 2(2): 1-9.