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# Analgesic effect of *Sansevieria longiflora* (Sim) water extract and xylocaine cream on surgical wound incision in rats

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## Abstract

Analgesic effect of *Sansevieria longiflora* (*S. longiflora*) on surgically created wound was studied. Phytochemical screening of the crude water extracts was carried out following standard procedures. *Sansevieria longiflora* leaves was heated and the sap was squeezed into a sample bottle to be used as it was traditional believed. Its water extract was also prepared following standard procedure. Four (4) albino rats consisting of 2 males and 2 females were used for the study. One centimeter full thickness skin incision was created on 4 different locations at the dorsal region of each rat under anaesthesia. Each of the 4 wounds created was topically treated once daily with *S. longiflora* sap (E), *S. longiflora* water extract (F), Physiological saline solution (C) and xylocaine cream (G). The treatment regimen was rotated in a clockwise direction on each rat. Pain perception was evaluated with an algometer at 6:00 GMT (morning) and 18:00 GMT (evening) from day 1 post surgery for 5 days. The phytochemical studies revealed that *S. longiflora* contains carbohydrates, glycoside, saponins, steroids, triterpenes, flavonoids, alkaloids and tannins. Day1 after the surgery, the mean morning value of force (N) required to elicit pain in xylocaine treated wounds ($5.2 \pm 0.79$) were significantly higher ($p<0.001$) than that of physiological saline solution ($2.8 \pm 0.29$) whereas, not different from that of *S. longiflora sap* ($4.48 \pm 0.57$) and water extract ($4.2 \pm 0.6$). Similarly, on the day 5 after the surgery, the mean morning value of force (N) required to elicit pain in xylocaine treated wounds were significantly higher ($p<0.0001$) than that of physiological saline solution (PSS) treated wounds and not different from that of *S. longiflora* sap and water extract. This showed that *S. longiflora* sap and water extract possess analgesic effect on incisional wounds. This study serves as a preliminary study on this plant towards searching for potent analgesics that could be used for procedural pain management.

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## Introduction

The medicinal properties of *Sansevieria* species is well documented (Wasciky and Hoehn, 1951; Mimaki *et al.,* 1996). This includes treatment of different types of pain conditions including colic and ear aches (Aliero *et al.,* 2008). The leaf sap could also be applied directly to infected wounds, cuts and grazes or used to treat fungal and scabies infection (Olivia, 2005). Traditionally, in treating ear aches and hemorrhoids, the leaves are heated and the warm juice is squeezed onto the affected area. In Nigeria, the leaves and roots of *Sansevieria liberica* are used in traditional medicine for the treatment of asthma, abdominal pains, colic, diarrhea, eczema, gonorrhea, hemorrhoids, hypertension, menorrhagia, piles, sexual weakness, snake bites and wounds of the foot (Osabohien and Egboh, 2008; Adeyemi *et al.,* 2009). Similarly, ethanol and water extracts of Sansevieria trifasciata leaves has been reported to show a dose-dependent and significant increase in pain threshold under different pain tests (Anbu *et al.,* 2009).

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**Fig. 1. Sansevieria *longiflora* (Sim)**

*Sansevieria longiflora* (Figure 1) is commonly referred to as Florida bowstring hemp, mother-in-law tongue, bowstring hemp. It is also known in Nigeria as *Oja ikooko* (Yoruba) or *Moda* (Hausa). Though it is commonly used as ornamental plants in offices along with *Sansevieria liberica* in various parts of the country, it is as well known for its medicinal purposes. In the North-Eastern and South-Western part of Nigeria, the leaf of *S. longiflora* is popularly used as ligature. It is strapped round a very deep cut from the cutlass, sickles or other sharp objects while working on the farm. Subsequently, the leave is heated so as to be able to squeeze out the sap into the wounds. This is traditionally used to manage deep wounds and chronic wounds that failed to heal. The root decoction is also used to treat whooping cough and diabetes in human.

Self-reporting system facilitates pain treatment in most human patients (Rocca *et al.,* 2015) unlike in animals. The research into pain assessment in animals is ongoing. Pain perception has been assessed by several authors in animals using different methods including analysis of physiological variables (heart rate, respiratory rate), measurement of hormones (cortisol, adrenaline) (Bufalari *et al.,* 2007), subjective pain scales (visual analogue scores, numerical rating scales) (Mathews *et al.,* 2014), analysis of electrophysiological parameters (electroencephalograms, evoked potentials), behaviour of the animal (Molony and Kent, 1997) and digital pressure palpation. Quantitative pain assessment could be done using equipments like digital force gauge algometer, anaesthesiometer and WAGNER Pain Test ™ Algometer. Pressure algometry has been proven to be a valid measure of localized pain in muscle, joints, tendons, ligaments and bones (Franco *et al.,* 2014; Manufacturer's guide). Hence, Pain Test ™ Algometer produced by WAGNER were used to test for pain threshold, tolerance and monitor the treatment progress by pressure algometry at different point of incision.

Stimulation of the peripheral neural receptors due to noxious stimuli like surgical incision leads to nociceptive pain (Woolf, 2010; Mark *et al.,* 2015). This leads to inflammatory pain which results gradually from the activation of the immune system in response to injury (Woolf, 2010; Mark *et al.,* 2015). Pain management is a key to success in medical practise (Mark *et al.,* 2015). Different modalities in managing pain are being discovered (Mathews *et al.,* 2014). However, several adverse effects are associated with the use of drugs. Respiratory depression, decreased gastrointestinal motility, increased appetite, sedation, euphoria and nausea are associated with administration of opioids (Coetzee, 2011) while gastric ulceration (Lascelles *et al.,* 2005), nephrotoxicity (Crandell *et al.,* 2004) and delayed bone healing (Ochi *et al.,* 2011) has been reported with Non-Steroidal anti-inflammatory drugs (NSAIDs). Hence, there is need for continual search of alternative agent that is more potent with little or no side effects. More so, phytotherapeautical agents are relatively cheaper and without fear of drug residue in the meat product (Dhama *et al*., 2015), hence this research becomes important. Since, *Sansevieria longiflora* is readily available and commonly used by traditional people in Nigeria to manage ailments related to pain, it is therefore, necessary to investigate the notion behind its usage.

## Materials and methods

*Collection and identification of the plant samples*

The leaves of the *S. longiflora* was collected from Obokun Local Government Area of Osun state Nigeria and was confirmed in the Herbarium of the Botanical Garden of Ahmadu Bello University, Zaria, with voucher specimen number of1821.

*Preparation of crude extract*

*Sansevieria longiflora* leaves was heated and the sap (E) was squeezed into a sample bottle to be used as it was traditional believed. To prepare the crude water extract of *S. longiflora* (F), the leaves were air dried and the dried powder of the plant was extracted and stored at 4° C in air tight bottles as described by Prashanthi *et al*. (2012).

*Phytochemical analysis*

The phytochemical screening of the crude extracts was carried out as described by Evans and Trease (1996), Harborne (1973) and Sofowara (1993) so as to evaluate for the presence of secondary metabolites such as saponins, alkaloids, flavonoids, steroids, tannins, cardiac glycosides, glycosides, and proteins.

*Experimental protocol*

Incisional wound model was adopted. Four albino rats consisting of 2 males and 2 females of 24 weeks old weighing about 200 ± 20 g were used. They were housed separately in a steel cage and fed with commercial rat feed (prepared from Vital Feed® grower mash) and water *ad libitum*. The research was conducted based on the ethical guideline and rules by the Ahmadu Bello University Committee on Animal Use and Care. The approval number given was ABUCAUC/2016/027. The animals were allowed to acclimatize for 2 weeks before the experiment. Each rat was anesthetized by intra muscular injection of Rompun(R) (xylazine hydrochloride) and ketamine hydrochloride at the dosage of 5 mg/kg and 40 mg/kg respectively. One centimeter full thickness skin incision was created on 4 different locations at the dorsal region of each rat under anaesthesia. Each of the 4 wounds created (E, F, C, and G) was topically treated once daily with *S. longiflora* sap, *S. longiflora* aqueous extract, Physiological saline solution (PSS) and Xylocaine cream respectively. The xylocaine cream consists of Lidocaine hydrochloride Gel B.P which was produced by Alves Healthcare Pvt. Ltd. Nangal Uperia, Swarghat Road, Road, India (Mfg. Lic No. HP/DRUGS/MB/07/676). The treatment regimen was rotated in a clockwise direction on each rat.

**Fig. 2. Evaluation of pain perception in rats using an algometer**

*Postoperative pain evaluation*

Pain perception was evaluated with an Algometer (digital force gauge machine: HF-200 produced by Orchid Scientific, B-59 MIDC, Ambad Nashik-422010 and product ID: ALGO.D.1/16.17/01) after 15 minutes of treatment at 12 hours intervals from day 1 post surgery for 5 days (Figure 2).

*Data analysis*

All data were expressed as means ± SEM (standard error of mean). Two ways ANOVA were used to analyze the data recorded. Bon ferroni post-hoc test was used for multiple comparisons. Statistical analyses were performed using the Graphpad prism 5.0 for sciences. P-Values of less than 0.05 were considered as statistically significant

**Results and discussion**

*Phytochemical studies*

The preliminary phytochemical studies (qualitative analysis) on the leaves of *S. longiflora* reveals that it contains carbohydrates, glycoside, saponins, steroids, triterpenes, flavonoids, alkaloids and tannins (Table I). Though there is limited information on the phytoconstituents of *S. longiflora* however, similar studies have revealed the presence of carbohydrates, glycoside, saponins, steroids, triterpenes, flavonoids and tannins in *Sansevieria roxburghiana* (Nweze *et al.,* 2004; Philip *et al.,* 2011; Pramoda and Hanumanth, 2015), *Sansevieria trifasciata* and *Sansevieria liberica* (Ogukwe *et al.,* 2004; Sunilson *et al.,* 2009). More so, *Sansevieria roxburghiana* was reported to contain quinone, proteins, anthocyanin and betacyanin (Nweze *et al.,* 2004; Philip *et al.,* 2011; Pramoda and Hanumanth, 2015).

*Analgesic properties*

The mean values of the force (N) required to eliciting pain increased with days in all the treated groups with slight variation as presented in the Figure 3 and 4. Generally the normal saline group tolerated less force (more pain) while the Xylocaine treated group tolerated more force (less pain). On the morning of day 1 after the surgery, the force (N) required in eliciting pain in *S. longiflora* sap, *S. longiflora* water extract, PSS and xylocaine cream were 4.48 ± 0.57, 4.2 ± 0.60, 2.8 ± 0.29, and 5.2 ± 0.79 respectively (Figure 3). While the force recorded in evening were 3.53 ± 0.86, 3.23 ± 0.36, 2.58 ± 0.33 and 5.40 ± 0.66 respectively (Figure 4). On the morning of day 5 after the surgery, the force (N) required to eliciting pain in *S. longiflora* sap, *S. longiflora* water extract, PSS and xylocaine cream were 9.83 ± 2.20, 6.93 ± 0.67, 5.93 ± 0.92 and 12.63 ± 3.78 respectively (Figure 3). While the force recorded in evening were 8.53 ± 1.19, 6.93 ± 1.22, 6.18 ± 0.90, and 9.95 ± 1.96 respectively (Figure 4).

On the morning of day 1 after the surgery, the maximum force required to elicit pain with xylocaine treated wound (5.2 ± 0.79) was significantly higher ($p < 0.001$) than that of PSS treated wounds (2.8 ± 0.29) whereas, not different from the force required by the *S. longiflora* sap (4.48 ± 0.57) and water extract (4.2 ± 0.6). While in the evening the force tolerated by the xylocaine cream treated wounds (5.4 ± 0.66) was significantly higher ($p < 0.001$) than that of PSS treated wounds

**Table I. Phytochemical constituents of *sansevieria longiflora***

| S/N | Chemical Test | *Sansevieria longiflora* |
|---|---|---|
| 1 | Carbohydrates | |
| | Molisch's test | + |
| | Fehling's test | - |
| 2 | Cardiac glycoside | |
| | Kella-killiani test | + |
| | Kedde's test | + |
| 3 | Free Anthraquinones | |
| | Borntrager's test | - |
| 4 | Combined Anthracene | |
| | Modified Borntrager's test | - |
| 5 | Saponins | |
| | Frothing test | + |
| | Hemolysis test | + |
| 6 | Steroids and Triterpenes | |
| | Leiberman-Burchards test | + |
| 7 | Flavonoids | |
| | Shinoda test | + |
| | Sodium hydroxide test | + |
| 8 | Tannins | |
| | Lead sub-acetate test | + |
| | Ferric chloride test | - |
| 9. | Alkaloids | |
| | Mayer's test | + |
| | Dragendorff's test | + |

Key: +: Present
- : Absent

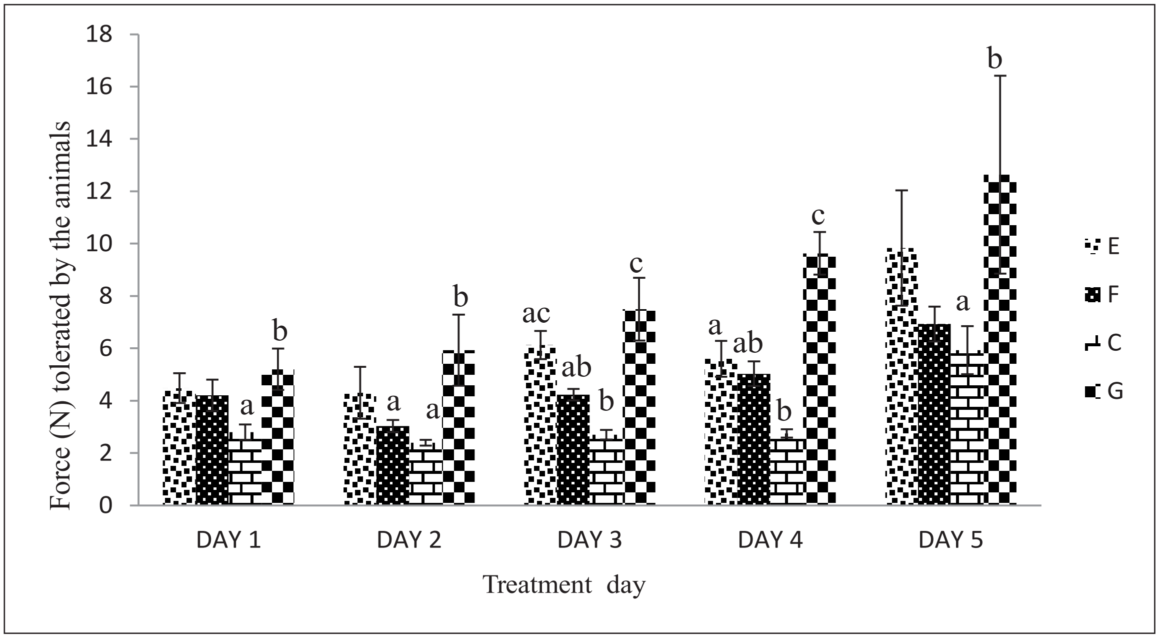


**Fig. 3. Mean value of maximum force (N) tolerated by the animals at different wound sites in the morning (n=4). KEY: E *(Sansevieria longiflora* sap); F (*Sansevieria longiflora* water extract); C (Physiological normal saline) and G (Xylocaine cream)**

[abc]Means in the same row with different superscript alphabets were significantly different (p<0.05)

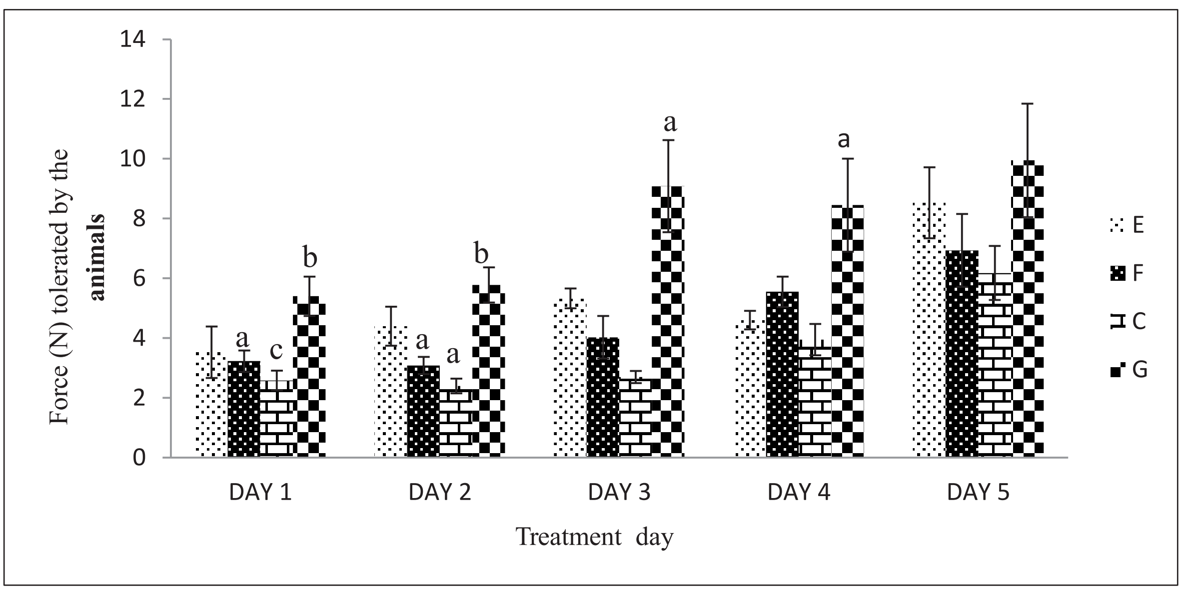


**Fig. 4. Mean value of maximum force (N) tolerated by the animals at different wound sites in the evening (n=4). KEY: E (*Sansevieria longiflora* sap); F (*Sansevieria longiflora* water extract); C (Physiological normal saline) and G (Xylocaine cream)**

[abc]Means in the same row with different superscript alphabets were significantly different (p<0.05)

(2.58 ± 0.33) and *S. longiflora* water extract (3.23 ± 0.36) and not different from that of *S. longiflora* sap (3.53 ± 0.86). This was similarly observed in the morning and evening of day 2 post incision. Whereas, on the day 3 and 4 post incision, the force tolerated by the xylocaine treated wounds was significantly higher than all other treatment groups, while on the morning of day 5 post incision, the force tolerated by the xylocaine treated wounds were significantly higher than that of PSS treated wounds and not different from that of *S. longiflora* sap and water extract. This showed that *S. longiflora* sap and water extract possess analgesic effect on incisional wounds however, the sap was more potent and effective than the water extract supporting the traditional methods of preparation. There is likelihood that some active ingredients were lost during the extraction.

This study has shown that *S. longiflora* possess some analgesic properties using the force gauge machine on experimental cutaneous wounds which was due to the flavonoids component of the secondary metabolites as described by Akpalo *et al.* (2015), Fakhim *et al.* (2015) and Christian *et al.* (2016). Flavonoids are free radical scavengers due to their high reactivity as hydrogen or electron donors. This explained their therapeutic potentials (Soobrattee *et al.,* 2005). Inflammatory condition is associated with pain. The development of inflammation is initiated by the release of mediators of different structures/classes including serotonin, histamine, cytokines and chemokines, nitric oxide (NO), reactive oxygen species (ROS), and lipid mediators, such as prostanoids, leukotrienes, and lipoxins (Verri Jr. *et al.,* 2012). Those derived from components present in the plasma include bradykinin and members of the complement and coagulation system (Robbins *et al.,* 2010). Flavonoids interfere with these molecules at various degrees, depending on the concentration/doses of flavonoids used *in vitro* and *in vivo* (Verri Jr. *et al.,* 2012). Studies have shown that flavonoids induce analgesic effects by inhibition of both inflammatory and neuropathic pain through mechanisms involving the inhibition of cytokine production (e.g., IL-1β), prostaglandin, nitric oxide production and via endogenous opioid dependent mechanisms (Verri Jr. *et al.,* 2012).

Flavonoids also inhibit the peroxidase active site of COX-1, COX-2, and 5 lipoxygenase (5-LO) (Burnett *et al.,* 2007), resulting in inhibition of prostanoids (prostaglandins and tromboxanes) and leukotrienes production, respectively. Morikawa *et al.* (2003) similarly reported that flavonoids inhibited the COX-2 peroxidase active site (Burnett *et al.,* 2007) as well as the nuclear factor kB (NF-kB) -dependent expression of COX-2. Inhibition of prostaglandins produced algesia as prostaglandins contributed to edema as a consequence of increased vascular permeability and modulated the activity of neurons responsible for painful stimuli perception and thermal regulation (Robbins *et al.,* 2010). However, COX-1 inhibitors were known for their increase in incidence of gastric and intestinal ulcerations while COX-2 inhibitors increased the risk of cardiovascular problems (Buerkle *et al.,* 2004). Since the activity of flavonoids do not rely exclusively on the inhibition of COX enzymes but affect other pathways concomitantly to COX inhibition, they do not present these common side effects of NSAIDs that target COX enzymes (Verri Jr. *et al*., 2012). In fact, flavonoids have opposite effects compared to the COX inhibitors. Hence, treatment with extracts containing flavonoids and/or flavonoid-rich fractions of extracts reduced the gastric lesions induced by NSAIDs, such as aspirin and indomethacin, or even promoted tissue healing (Alam *et al.,* 2010; Vasconcelos *et al.,* 2010).

Anbu *et al.* (2009) reported dose-dependent analgesic effects of ethanol and water extracts of Sansevieria trifasciata leaves. Jimuty *et al.* (2012) similarly reported the analgesic, cytotoxic and antioxidant activity of the whole plant part of *S. roxburghiana*. Other studies on *Sansevieria* species have documented its anti inflammatory activity (Da Silva *et al.,* 2003) and analgesic property (Sunilson *et al.,* 2009).

## Conclusion

This study has revealed that *S. longiflora* sap and water extract has analgesic effects on incisional wounds which was due to the flavonoids contents of the secondary metabolite. This explains the use of this plant by the traditional medicine practitioner in Nigeria for different ailment relating to alleviation of pain. This study will serve as a preliminary study on this plant towards searching for potent analgesics for post procedural pain management.

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