Evaluation of Wound Healing Activity of Salvia Hypoleuca Extract on Rats

Jasem Estakhr*, Nasim Javdan

Science and Research Branch, Islamic Azad University, Fars, Iran.

***Corresponding Address:** Jasem Estakhr, j.estakhr@yahoo.com. Tel: +989179283966.

Summary

The extract of Salvia hypoeuca was evaluated for their wound healing activity in Albino Wistar rats using chemically induced wound model. The experimental animals were chemically burned by conc HCL (80%), and then divided into four groups, control, standard (Nitrofurazone 0.2% w/w), low dose (150 mg/kg) and high dose (300mg/kg). The re-epithelialization was evaluated by the reducing width of wounds for seven days. It was observed that the alcoholic extract showed re-epithelialization faster when compared with control. The extract also showed significant wound healing activity when compared with standard. Results from this study support that the use of alcoholic leaf extract of Salvia hypoleuca in topical skin care and healing wounds.

Keywords: Salvia hypoleuca, Wound healing activity, Rat.

Introduction

The plants of the genus Salvia, which consist about 900 species (1) are generally known for their multiple pharmacological effects such as analgesic and anti-inflammatory (2), antioxidant (3), hepatoprotective (4), hypoglycemic activities (5), and antiischemia (6, 7). The results of recent studies have shown that some species of Salvia impress fertility in rat. The data obtained from previous studies clearly demonstrated that Salvia hematodes enhances the orientation of males towards the female by increased anogenital behavior and enhance licking and grooming of the genitals (8). This plant also increases the ejaculation latency. Salvia fruticosa produces adverse effects on the fertility of male and female rats (9). Estakhr and Javdan (2011) have demonstrated the existence of phenolic compounds (SH1, 3, 2 and 4) i.e. Gallic acid and ellagic acid (phenolic acids), rutin and quercitin (flavonoids) in the ethanolic extract of Salvia hypoleuca (10).The presence of these constituents may be one of the contributing factors responsible for the activities by virtue of their different properties like anti oxidant, anti inflammatory, analgesic and antimicrobial activities. Several phytoconstituents like triterpinoids, (11) tannins, saponins(12),alkaloids(13) and flavonoids(14) are known to promote wound healing process due to their antioxidant and antimicrobial activities. Since there is hardly any work reported on the effect of Salvia hypoleuca on wound, the present study was planned to evaluate effects of Salvia hypoleuca on wound using rats as an experimental animals.

Materials and Methods

Plant materials

Physalis alkekengi was collected from Guilan province, and then was identified by a botanist. Fresh plant was thoroughly washed using deionized water, separated into leaves and fruits, and mopped with tissue paper and air-dried in shade so as to prevent the decomposition of chemical constituents. One gram of the material was ground into fine powder using blender. Hundred gm of powder was suspended in 250 ml of distilled water and allowed it to stand overnight in refrigerator; it was then sieved through several layers of muslin cloth. The filtrate (water extract) was discarded. The residue was extracted with 95% ethanol using sox halation wherein ethanol was evaporated in a rotatory evaporator at 40–50 ºC. The yield was 3.2 g/100 g of powder.

Animals

Eighteen male albino rats of 6–8 weeks age, 150–200 gm of weight were procured from Razi Institute, (Karaj, Iran). The animals were housed in well ventilated, air conditional animal house at a constant temperature of 23±20c, with the relative humidity of 55-60%. The animals were housed in spacious polypropylene cages with paddy husk as bedding material. The animals were maintaining on standard pellet diet and purified water. The animals were provided with food and water and libitum except during fasting. Re-epithelisation time was observed for 1 to7 days and measured wound healing diameter.

Induction of wound and treatments

The skin of rats were shaved mechanically and leaved for 24hours, two or three drops of concentrated HCL (80%) were topically applied carefully on the shaved skin. Then the skin burned rats were housed separately under sterile conditions in isolated room cleaned with sterile solution. The two experimental groups (low dose (150mg/kg body weight) and high dose (300mg/kg body weight)) were treated twice a daily by extract of Salvia hypoleuca. One of these groups treated by 0.2%w/w Nitrofurasone USP (Standard), while the others are treated with vehicle (20% Tween 80) treated as Control. The areas of the burns were recorded daily to show the contracting ability of wounds and to determine the closure time. The granulations of tissues or rebuilt layers were observed. The re-epitheliaisation time was recorded.

Statistical analysis

Results were analyzed by one way ANOVA followed by Dunnet multiple comparisons tests. P values <0.05 were considered as significant.

Results

The table 1 shows the Mean ± SE values of wound diameter of four groups. There was a statistically significant ($p<0.01$) reduction in the mean wound diameter, when comparing Salvia hypoleuca 150 mg/kg body weight and 300 mg/kg body weight with Nitrofurazone 0.2% w/w. The figure 1 indicates that the extract showed significant wound healing activity at the dose of 300mg/kg body weight than 150mg/kg body weight.

Table 1: Effect of Salvia hypoleuca extract on wound diameter in low and high dose.
ns p>0.05, *p<0.05, ** p<0.01 – one way ANOVA followed by Dunnet multiple comparisons tests

Groups	Day I	Day II	Day III	Day IV	Day V	Day VI	Day VII
Control	10±0.35	9.63±0.074	9.64±0.123	9.17±0.023	8.7±0.143	8.3±0.023	7.8±0.123
High Dose	10 ± 0.53^{ns}	$8.1\pm0.054^{**}$	$6.2\pm0.73^{**}$	$5.3\pm0.62^{**}$	$4.5\pm0.52^{**}$	$3.9\pm0.32^{**}$	$2.6\pm0.42^{**}$
Low Dose	10.01 ± 0.21^{ns}	$8.2\pm0.12^{**}$	$7.3\pm0.82^{**}$	$6.4\pm0.47^{**}$	$5.7\pm0.33^{**}$	$4.85\pm0.12^{**}$	$4.1\pm0.32^{**}$

Discussion

In the present study the wound healing activity of alcoholic extract of Salvia hypoleuca was evaluated by chemically induced wound model in Albino Wistar rats. Several phytoconstituents like triterpenoid, tannins, saponins, alkaloids and flavonoids are known to promote wound healing process due to their antioxidant and antimicrobial activities. In addition triterpenoid reported to possess an ability to increase the collagen content, which is one of the factors promoting wound healing. Estakhr and Javdan (2011) have demonstrated the existence of phenolic compounds (SH1, 3, 2 and 4) i.e. Gallic acid and ellagic acid (phenolic acids), rutin and quercitin (flavonoids) in the ethanolic extract of Salvia hypoleuca (10). Wound healing involves different phases such as contraction, epithelialization, granulation and collagenation(15). The decreased wound diameter significantly contributes to better and effective wound healing property. During the initiation of study from "0" there was not much different in healing of wounds in all the four groups. At the end of the study (7th day) - low dose and high dose showing significant wound healing effect. When the results were interpreted in high dose, showed high significant wound healing activity as compare to low dose. High dose showed better and faster wound healing activity as compare to low dose. The induction wound study after treatment with alcoholic extract of Salvia hypoleuca on 7th day, regenerated tissue shows increase in tensile strength as compared to untreated group. Standard and extract of Salvia hypoleuca treatment increase the tensile strength and significant wound healing activity.

References

1. Brickell C. Encyclopedia of garden plants. London: Dorling Kindersley; 1996, p. 926.
2. Hernandez-perez M, Rabanal RM, de la Torre MC, Rodriguez B. Analgesic, antiinflammatory, anti pyretic and haematological effect of aethiopinone, an onaphthoquinone diterpeniod from Salvia anthiopis roots and two hemisynthetic derivatives. Planta Med 1995; 61:505–9.
3. Cuppett SL, Hall CA. Antioxidant activity of the Labiatae. Adv Food Nutr Res 1998; 42:245–71.
4. Wasser S, Ho JM, Ang HK, Tan CE. Salvia miltiorrhiza reduce experimentallyinduced hepatic fibrosis in rats. J Hepatol 1998; 29:760–71.
5. Jimenez J, Risco S, Ruiz T, Zarzuelo A. Hypoglycemic activity of Salvia lavandulifolia. Planta Med 1996; 4:260–2.

6. Akbar S, Tariq M, Nisa M. A study on CNS depressant activity of Salvia haematodes wall. Int J Crude Drug Res 1984; 22:41–4.
7. Yu WG. Effect of acetylsalvianolic acid A on Platelet function. Yao Xao Xue 1994;29:412–6.
8. Islam MW, Tariq M, Ageel AM, Al-said MS, Al-yahya AM. Effect of Salvia haematodes on sexual behavior of male rats. J Ethnopharmacol 1991; 33:67–72.
9. Elbetieha A, Al-hamood MH, Alkofahi A, Bataineh H. Reproductive Toxicity potentials of Salvia fruticosa (Labiatae) in rats. J Ethnopharmacol 1998; 61:67–74.
9. Nasim J., Jasem E. Isolation of phenolic compounds from the ethanolic extract of Salvia hypoleuca. Pharmacologyonline, 2011; (in press).

11. Somava.L.I., Shode F.O., Ramnanan P.and Nadar A., Antihypertensive atherosclerotic and anti oxidant activity of triterpenoid isolated from olea europaea,subspecies Africanaleaves. J. Alcoholicpharmacol., 2003; 84, 299.
12. Gulcin I., Mshvidadze V.,Gepdiremen A and Elias R., Antioxidant activity of saponins isolated from lvy : alpha-hederin, hederasaponin-C,hedera calchiside –E and hedera calchiside-F. Plant medica, 2004; 70(6), 551.
13. Marjore M.C., plant product as antimicrobial agent. Clinical microbiology reviews, 1999; 12(4) 564.
14. T.suchiya H.,sato M.,Miyazaki.,Fujiwara S.,and Linum M., comparative study on antibacterial activity of phytochemical flavonones against methicillin resistant Staphylococcus aureus. J. Ethanopharmacol., 1996; 50, 27.

15. Purna K.S., Neelakanta P.R., and Babu M., investigations on wound healing by using amphibian skin. Indian. J. Exp. Biol.,1995; 33, 673-676.