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HEALING ACTIVITY OF *LEPIDIUM SATIVUM* L. IN EXPERIMENTALLY INDUCED DIABETIC RATS.

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ABSTRACT

Aim: The aim of present study was to evaluate the wound healing potential of *Lepidium sativum* seeds when administered orally as well as topically in STZ induced diabetic rats. **Methods:** Excision wound model was used in Sprague dawley rats of either sex. For assessing the wound healing property ethanolic extract of seeds was prepared and formulated into 20 and 30 mg/kg solution in water and also as 2% ointment in Vaseline. Control animals were treated with water as vehicle and Vaseline topically. The parameters studied include wound area measurement, % wound healing, Hydroxyproline and Hexosamine content along with some biochemical parameters. **Results:** Significant wound healing activity was observed both topically and orally treated groups. Ethanolic extract treated groups showed a significant increase in % wound healing, hydroxyproline and hexosamine content, RBC & WBC count and total protein content. Whereas consequently decreased wound area, clotting time, total cholesterol and serum triglycerides levels. For wound healing property higher oral dose (30 mg/kg) was found to be more effective than other and its observations were comparable with those of standard (mupirocin). **Conclusion:** The data of this study indicates that ethanolic seed extract of *Lepidium sativum* is effective in treatment of diabetic wounds. This property of the plant may be attributed to its antihyperglycaemic and anti-inflammatory property.

KEYWORDS: *Lepidium sativum seeds, excision wound model, hexosamine, hydroxyproline.*

INTRODUCTION

Diabetes mellitus is one of the most common chronic diseases in nearly all countries, and continues to increase in numbers and significance, as economic development and urbanization lead to changing lifestyles characterised by reduced physical activity, and

increased obesity.[1] Diabetic complications are becoming serious health issues these days, one of which is impaired wound healing.[2] Healing impairment in diabetes is characterized by delayed cellular infiltration and granulation tissue formation, decreased collagen organization, diminished blood flow, increased blood viscosity and reduced angiogenesis. [3,4,5] A wound occurs when the integrity of any tissue is compromised. The response to injury is immediate and the damaged tissue or wound then passes through phases in order to affect a final repair. [6] Wound healing is a physiologic process involving a series of sequential yet overlapping stages. There are anywhere from 3 to 5 stages of wound healing, depending on how the various biologic mechanisms are linked. The first stage, haemostasis, occurs immediately at the time of injury and is usually completed within hours. The second stage, inflammation, begins shortly after haemostasis and is usually completed within the first 24 to 72 hours after injury4; however, it may last as long as 5 to 7 days after injury. Proliferation and repair, the third stage, typically occurs 1 to 3 weeks after injury. The fourth and final stage, remodelling, begins approximately 3 weeks after injury and may take anywhere from months to several years to achieve physiologic completion. Thus, although the skin seems intact within days to several weeks after an injury, the tissue underneath is still vulnerable to damage as it undergoes the final stages of wound healing. [7] Plants are still a major source of medicines. The use of traditional plant extract has been flourished recently. *Lepidium sativum Linn*. (family: Cruciferae) is commonly known as "Common cress", "Garden cress" or "Halim". It is a small smooth erect annual herb and is of flavonoid (2.50%) and saponin (2.96%), whereas the percentage yields of alkaloid and total phenol recorded were minimal (0.29 and 0.94, respectively).[9] *Lepidium sativum* has potential as foodstuff as well as neutraceutical. Traditionally its seeds are known to be effective against diabetes, hypertension, kidney stones, inflammation, bronchitis, rheumatism, muscular pain as well as in treating cancer. LS is known to increase breast milk after pregnancy. [10] The scientifically proven potent antioxidant,[11] anti inflammatory [12] and hypoglycaemic [13] properties of *Lepidium sativum* seeds makes it a suitable source for a new target of wound healing in diabetic condition. Literature survey reveals that till date no such study has been carried out on this herb.

MATERIALS AND METHODS

Plant materials

The dried seeds of *Lepidium sativum* were purchased from local ayurvedic drug shop of Pune, Maharashtra. These seeds were authenticated by Botanical Survey of India, Pune.

Plant extraction and standard used

The seeds of *Lepidium sativum* were dried completely and grinded to fine powder. Then powder of *L. sativum* was defatted with petroleum ether (40–60%) until free from fatty material, and then extracted thoroughly with ethyl alcohol (95%) by maceration. The alcohol from the extract was removed under reduced pressure and finally dried in desiccators. The dried ethanolic extract was further used for estimating the wound healing activity.

Experimental animals

Healthy Sprague dawley rats of either sex weighing between 150-200 gm were procured from National Institute of Biosciences, Pune. Animals were kept in animal house of Sinhgad College of Pharmacy, Vadgaon, Pune. They were provided standard laboratory diet (Amrut feed, India) and clean drinking water *ad libitum*. Animals were housed in group of six in polypropylene cages. They were maintained at controlled temperature (23 ± 20 C), humidity (55 ± 5%) and 12 hrs dark and light cycle. All the procedures in this study were approved by institutional animal ethics committee, (SCOP/IAEC/2012-13/12), constituted for the purpose of control and supervision of experimental animals by Ministry of Environment and Forests, Government of India, New Delhi, India.

Chemicals

Streptozotocin (STZ) (Enzo Life Science, Exeter, UK), Petroleum ether (Loba Chemie, Mumbai, India), Ethanol (Analab fine chemicals, Mumbai, India), Vaseline ointment (Hindustan lever ltd, Andheri (E) Mumbai, Maharashtra), Mupirocin ointment (GSK, Nasik, India), Hydroxyproline (Sigma Chemicals, St. Louis), Glucosamine HCl (Otto Chemika, India), Chloramine-T (SD Finechem, Mumbai, India), Acetyl acetone (Loba Chemie, Mumbai, India) and other diagnostic kits (Biolab diagnostics, Mumbai, India) were used in this study. The drugs solutions were prepared freshly before administration on daily basis and stored in amber coloured bottles.

Dose of drug

The dose of extract of *Lepidium sativum* was selected based on the earlier studies [13] in which the 20 mg/kg was proved to be effective as anti hyperglycaemic. Hence the doses selected are 20mg/kg and 30mg/kg of *Lepidium sativum* ethanolic extract orally and an ointment of concentration 2% was applied topically in accordance with the mupirocin ointment which is used as a standard.

Blood collection

Blood was collected by puncturing retro-orbital plexus of rats under mild ether anaesthesia using fine glass capillary in epindorff tubes. The blood was allowed to clot at room temperature and serum was separated by centrifugation at 3000 rpm (g=506.11) for 10 min.

Induction of diabetes

All the rats were fasted overnight before administration of streptozotocin (STZ). STZ will be prepared in citrate buffer (pH 4.5, 0.05 M). Streptozotocin (60 mg/kg, i.p.) was given to different groups of rats. Control rats were injected with citrate buffer only. 2 days after STZ injection, blood samples were collected, and plasma glucose levels estimated by using Glucometer. After a week, surviving rats with fasting serum glucose level higher than 250 mg/dl were used for the study.[14]

Excision wound model

Excision wound model was used for the study of rate of contraction of wound and epithelization. Animals were anaesthetized with 80 mg/kg dose of ketamine (i.p.) and the back hairs of the animals were depilated by shaving. An impression was made on the dorsal thoracic region on the anaesthetized rat. Excision wounds sized 3.14 cm^2 (1 cm radius) were made by cutting out layer of skin from the shaven area. Haemostasis was achieved by blotting the wound with cotton swab dipped in normal saline. [15]

The study comprised of seven different groups, each group consisting of 6 animals as follows:

Group I (NC) served as normal control and were given only water. Group II (VC) was Vaseline control whose wounds were applied with plain Vaseline ointment. Groups III to VII were diabetic. Group III (DC) served as diabetic control and was administered with distilled water. Gruop IV (LS 20) and V (LS30) animals were administered orally with *Lepidium sativum* extract solution in water at a dose of 20 mg/kg and 30 mg/kg repectively. Group VI (LS 2%) animals were applied with *Lepidium sativum* ointment (2%) and Group VII (mupirocin) was standard group and was applied with mupirocin (2%) ointment.

Wound evaluation

The wounds were monitored and the area of wound was measured on 4, 6, 8, 10, 12, 14, 16 post-wounding days. The mean % wound closure is reported Wound healing rate was expressed as:

$$\% \text{ of wound closure} = \frac{\text{wound area on day 0} - \text{wound area on day n} \times 100}{\text{wound area on day 0}}$$

Where n =number of days 4th, 8th, 12th, 16th day15.

Biochemical markers determination

Initially the granulation tissue formed over the wound was removed on 4th, 8th, 12th and 16th day after wound creation. This tissue is first dried in hot air oven to constant weight and then used for further estimations.

Hydroxyproline estimation

The dried tissue was hydrolysed with 6N HCl at 1200c for 2-4 hrs in an autoclave. The hydrolysed samples were dried in desiccators and buffer was added to dilute samples to assay sensitivity range. Chloramine T reagent was added to samples and allowed to react for 20 min, then aldehyde perchloric acid reagent was added to the samples and samples were placed in waterbath (600c) for 15 mins. Following that the samples were cooled and absorbance was read at 550 nm on a UV/Vis spectrophotometer (shimadzu, Columbia, Maryland 21046 U.S.A). [16]

Hexosamine estimation

The tissue already dried was hydrolysed with 6 N HCl at 98ºC for 8 hrs. This hydrolysate was neutralised to pH7 with 4 N NaOH and then diluted using distilled water. Acetylacetone solution was added to above solution and heated to 96ºC for 40 min. This mixture was cooled and 96% ethanol was added, followed by pdimethylamino- benzaldehyde solution (Ehrlich's reagent). The above solution was mixed thoroughly and kept for 1 hr at room temperature. After an hour the absorbance was measured using a UV/Vis spectrophotometer (shimadzu, Columbia, Marylan 21046 U.S.A). [17] The amount of hydroxyproline and hexosamine were determined by comparing with the respective standard curves. The contents of both were expressed as mg/g dry tissue weight.

Estimation of clotting time

The tail vein of animal was sterilized with ethanol and then bold prick was made to have free flow of blood, blood was sucked in capillary glass tube of 15 cm long. A small bit of glass tube was carefully broken of every 15 seconds until a fine thread of clotted blood appeared. The period in between appearance of blood in tail and formation of clot is taken as clotting time.

Statistical Analysis

Values are expressed as Mean ± SEM. Results were analyzed using two-way ANOVA followed by Bonferroni test. The values were considered to be statistically significant at $p < 0.05$.

RESULTS

Blood glucose level

There was significant increase in blood glucose level in diabetic wound control rats when compared with normal wound control rats ($p< 0.001$). However, treatment with LS 20 and LS 30 for 16 days significantly decreased the blood glucose level as compared to diabetic wound control ($p<0.001$). (Table 1).

Effect of *Lepidium sativum L.*extract on blood glucose level in STZ induced diabetic rats

There was significant increase in blood glucose level in diabetic wound control rats when compared with normal wound control rats ($p< 0.001$). However, treatment with LS 20 and LS 30 for 16 days significantly decreased the blood glucose level as compared to diabetic wound control ($p<0.001$).

Table 1: Effect of *Lepidium sativum L.*extract on blood glucose level in STZ induced diabetic rats

Group	Blood glucose level (mg/dl)	
	Day 0	Day 16
NC	87.50±2.23	83.66±3.03
VC	88.83±1.77	81.00±2.68
DC	349.50±14.21*	322.16±11.96*
LS(20)	428.83±8.70	150.33±11.54#
LS(30)	437.16±7.06	98.33±3.47#
LS 2%	436.16±11.68	400.883±6.82
Mupirocin 2%	350.50±7.84	378.33±6.28

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC): Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS 30): *Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically.

n = 6, values are mean ± SEM*$p< 0.001$ as compared to NC group and #$p<0.001$ as compared to DC group on respective days**.** Statistical analysis was done by using two way ANOVA followed by Bonferroni test.

Clotting time

Clotting time was significantly increased in diabetic wound control rats as compared to normal wound control ($p<0.001$). Whereas treatment with LS 20 and LS 30 for 16 days significantly decreased clotting time when compared with diabetic wound control rats ($p<0.001$). (Figure 1)

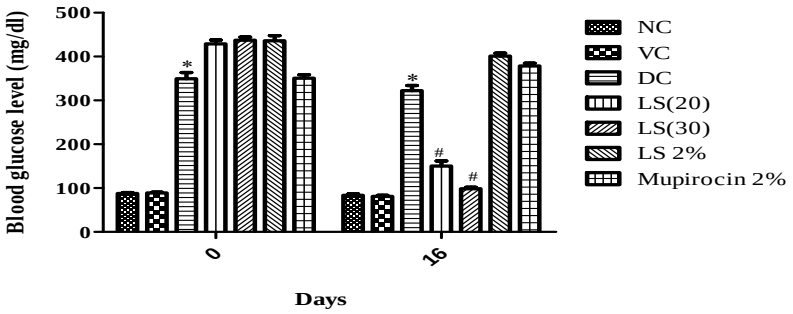


Fig 1: Effect of *Lepidium sativum L.* on blood glucose level in STZ-induced diabetic rats

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC): Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS 30): *Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically. n=6, values are mean ± SEM*$p<0.001$ as compared to NWC group and #$p<0.001$ as compared to DWC group on respective days .Statistical analysis was done by using two way ANOVA followed by Bonferroni test.

Hydroxyproline content

There was significantly decreased hydroxyproline content was observed in diabetic wound control as compared to normal wound control ($p<0.001$). However orally as well as topically treated LS groups shows significantly increased in hydroxyproline content when compared to diabetic wound control rats ($P<0.001$).(Figure 2).

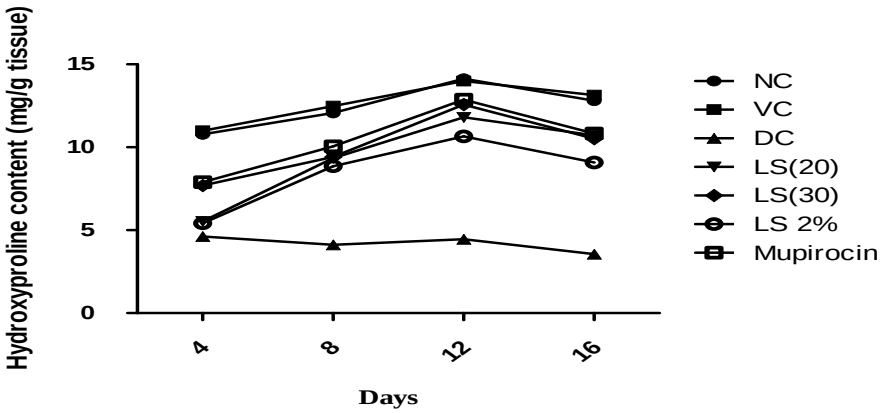


Fig 2 : Effect of *Lepidium sativum L.* extract on hydroxyproline content in STZ-induced diabetic rats

n=6, values are mean ± SEM. $^{*}p<0.001$ as compared to NC group and $^{@}p<0.05$,$^{\$}p<0.01$, $^{\#}p<0.001$ compared to DC group on respective days. Statistical analysis was done by using two way ANOVA followed by Bonferroni test.

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC): Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS 30): *Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically.

Hexosamine content

Hexosamine content was significantly decreased in diabetic wound control as compared to normal wound control ($p<0.001$). However orally and topically treated LS groups shows significantly increased hexosamine content when compared with diabetic wound control rats ($P<0.001$).(Figure 3).

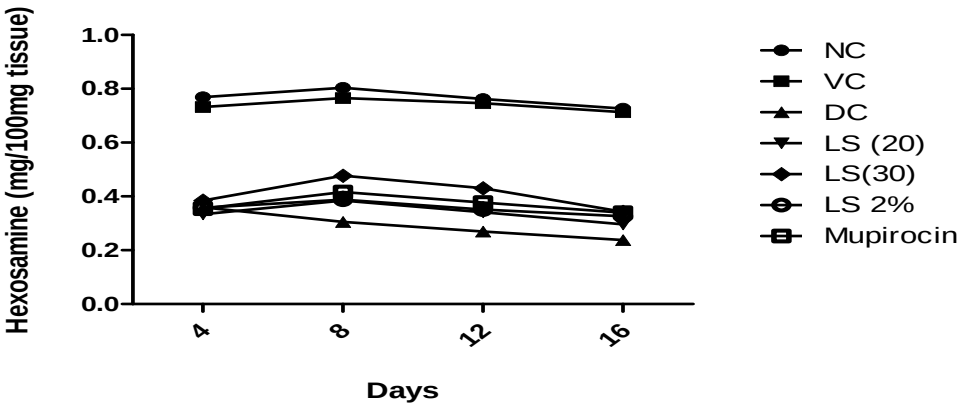


Fig 3: Effect of *Lepidium sativum L.* extract on hexosamine content in STZ-induced diabetic rats

n=6, values are mean ± SEM *p<0.001 as compared to NC group and $^{@}$p<0.01, $^{\#}$p<0.001 compared to DWC group on respective days. Statistical analysis was done by using two way ANOVA followed by Bonferroni test.

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC): Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS 30): *Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically.

Wound area

There was significant increase in wound area was observed in diabetic wound control when compared with normal wound control (p<0.001). However, orally and topically treated LS showed significant decrease in wound area when compared with diabetic wound control rats (p<0.001). (Table 2).

Table 2: Effect of *Lepidium sativum L.* extract on wound area in STZ-induced diabetic rats

Group	Wound area (cm^2)				
	Day 0	Day 4	Day 8	Day 12	Day 16
NC	3.08±0.05	2.60±0.03	2.20±0.16	1.47±0.01	0.73±0.03
VC	3.10±0.04	2.69±0.04	2.28±0.03	1.61±0.01	0.86±0.01
DC	3.11±0.05	2.88±0.02$^{@}$	2.53±0.14$^{\$}$	1.83±0.01$^{\$}$	0.97±0.04$^{\&}$
LS(20)	3.03±0.06	2.33±0.01*	1.56±0.01*	0.88±0.02*	0.43±0.03*
LS(30)	3.02±0.04	2.17±0.01*	1.25±0.01*	0.55±0.02*	0.22±0.01*
LS 2%	3.10±0.02	2.63±0.07$^{\#}$	1.20±0.01*	0.77±0.01*	0.42±0.03*
Mupirocin 2%	3.11±0.03	2.29±0.17*	1.37±0.05*	0.45±0.02*	0.18±0..02*

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC): Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS 30): *Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically. n = 6, values are mean ± SEM @p<0.01 as compared to NC group and #p<0.05, $p<0.01, *p<0.001 compared to DC group on respective days Statistical analysis was done by using two way ANOVA followed by Bonferroni test.

%wound closure

There was significant decrease in % wound closure was observed in diabetic wound control when compared with normal wound control (p<0.001). However, orally and topically treated

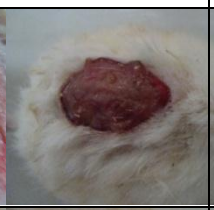
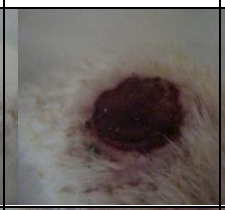
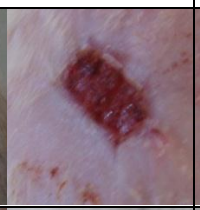
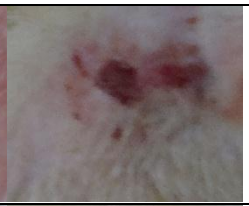
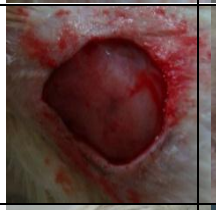
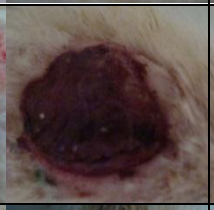
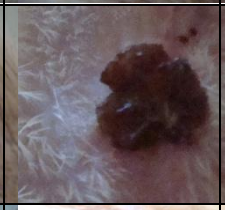
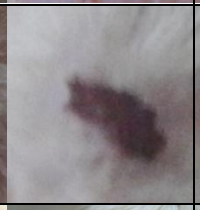
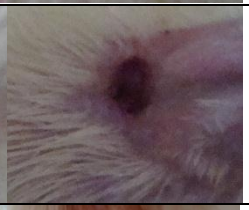
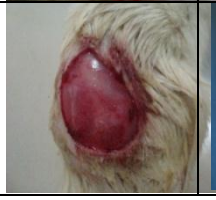
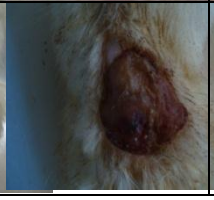
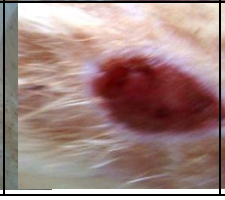
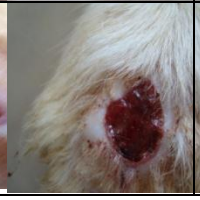
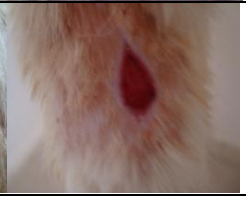
LS for 16 days showed significant increase in % wound closure when compared with diabetic wound control rats ($p<0.001$) (table 3).

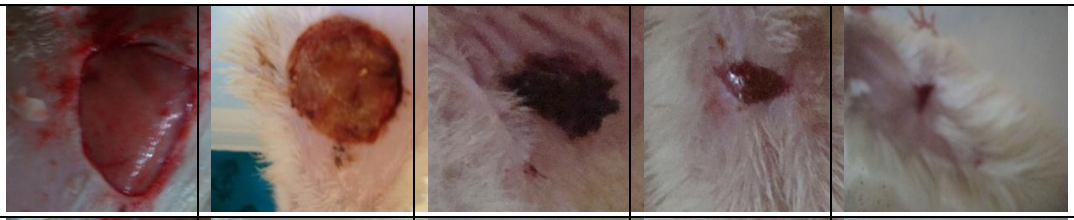
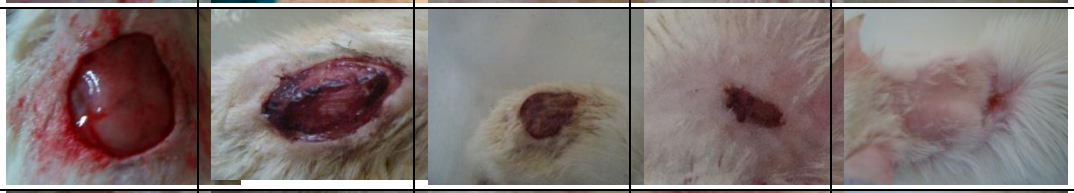
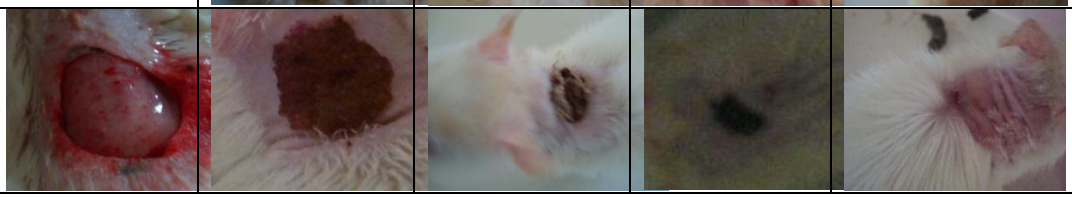
Table 3: Effect of *Lepidium sativum L.* aq.extract on % wound closure in STZ-induced diabetic rats

Group	% Wound closure			
	Day4	Day 8	Day 12	Day 16
NC	15.79±1.01	34.13±3.99	52.38±0.59	75.43±1.41
VC	13.37±1.30	26.35±0.96	48.12±0.47	72.28±0.61
DC	7.97±1.20$^{@}$	24.01±2.32$^{@}$	41.23±0.45$^{@}$	68.81±1.39 $^{\$}$
LS(20)	23.31±0.67*	48.68±0.53*	70.97±0.68*	85.79±1.14*
LS(30)	27.91±0.35*	58.50±0.60*	81.75±0.67*	92.62±0.59*
LS 2%	14.96±2.32$^{\#}$	57.37±3.76*	75.11±0.45*	86.32±1.02*
Mupirocin 2%	31.51±0.39*	55.78±1.61*	85.04±0.83*	93.98±0.64*

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC)Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS30):*Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically. n=6, values are mean ± SEM. @$p<0.05$, \$$p<0.01$ as compared to NC group and #$p<0.01$, *$p<0.001$ compared to DC group on respective days. Statistical analysis was done by using two way ANOVA followed by Bonferroni test.

Photograph 1: Photographs of excision wound on day 0, 4, 8, 12, & 16

GROUP	DAY0	DAY4	DAY8	DAY12	DAY16
NC					
VC					
DC					

LS (20)					
LS (30)					
LS 2 %					
MUPIROCIN					

Group I (NC): Normal control group, Group II (VC): Vaseline control group, Group III (DC): Diabetic control, Group IV (LS 20): *Lepidium sativum* 20 mg/kg orally, Group V (LS 30): *Lepidium sativum* 30 mg/kg orally, Group VI (LS 2%): *Lepidium sativum* ointment 2% topically, Group VII (Mupirocin 2%): Mupirocin ointment topically.

DISCUSSION

Diabetes is a group of chronic diseases characterized by hyperglycemia. Modern medical care uses a vast array of lifestyle and pharmaceutical interventions aimed at preventing and controlling hyperglycemia. In addition to ensuring the adequate delivery of glucose to the tissues of the body, treatment of diabetes attempts to decrease the likelihood that the tissues of the body are harmed by hyperglycemia. The direct and indirect effects on the human vascular tree are the major source of morbidity and mortality in both type 1 and type 2 diabetes. Generally, the injurious effects of hyperglycemia are separated into macrovascular complications (coronary artery disease, peripheral arterial disease, and stroke) and microvascular complications (diabetic nephropathy, neuropathy, and retinopathy).[18] Diabetes mellitus affects about 150 million people worldwide, and may be one common chronic noninfectious diseases with slowly course and symptomless. Diabetes has also been shown to affect cell and tissue metabolism in different ways, and many studies have demonstrated these effects in association with the chronicity of the disease.[19] Diabetic

individuals exhibit a documented impairment in the healing of acute wounds. Impaired wound healing is a serious complication of diabetes, and precedes 84% of all diabetics. The impaired healing in persons with diabetes involves multiple complex pathophysiological mechanisms. [20]

Wound is one in which the skin or another external surface is torn, pierced, cut, or otherwise broken. Contusions, small incisions, and abrasions tend to be non threatening, though some may pose the risk of infections. Skin-wound healing is an orchestrated biological phenomena consisting of three sequential phases, inflammation, proliferation, and maturation. Many biological substances are involved in the process of wound repair, and this short and simplified overview of wound healing can be adopted to determine wound vitality or wound age in forensic medicine. With the development of genetically engineered animals, essential molecules for skin wound healing have been identified. Especially, cytokines, and growth factors are useful candidates and markers for the determination of wound vitality or age. Moreover, bone marrow-derived progenitor cells would give significant information to wound age determination. In this review article, some interesting observations are presented, possibly contributing to the future practice of forensic pathologists.[21]

In the present study, the effect of *Lepidium sativum* (oral and topical) and standard drug mupirocin (topical) in diabetic wound healing was studied in STZ induced diabetic rats using excision wound rat model. This is the first report to show that *Lepidium sativum* accelerates the wound healing process in diabetic animals. Experimental diabetic rats exhibited decreased collagen However LS treatment resulted in improvement in hematological parameters, inhibition of lipid peroxidation, greater collagen deposition, and hence improved wound healing. In the present study intraperitoneal injection of STZ (70 mg/kg) produced significant elevation in serum glucose level and produced hyperglycemia which is in agreement with previous study. [22] STZ-induced diabetic rats exhibited hyperglycemia and decrease in body weight. The present study has demonstrated diminutions in serum glucose concentration and an increase in the body weight in diabetic rats treated with *Lepidium sativum* (20 and 30 mg/kg) without affecting the insulin release. This could be because of presence of alkaloids. The exact mechanism of reducing blood glucose level is not yet known.[9,13] Healing requires collaborative effect of various tissues and cells in a stepwise manner. It includes platelet aggregation, blood clotting, formation of fibrin, and inflammatory response to injury, angiogenesis and reepithelialization. [23] Calcium and vitamin K are clotting factors which are

depleted in diabetic condition. These both factors work together along with fibrinogen in clot formation. Thus without sufficient avaibility of clotting cascade delayed clot formation occurs. [24] In the present experiment animal groups treated with LS orally (20 and 30 mg/kg) showed increased clotting time than diabetic control animals.Collagen is one of the more important structural proteins in the body being of particular importance in connective tissues by providing their durability. Collagen is one of the few proteins which contain the amino acid hydroxyproline. [16] There is an in vivo pathway through which newly synthesized collagen when degraded releases free hydroxyproline and some of its peptides. The source of these degradation products is procollagen, and the degradation site is intracellular, and this pathway is greatly enhanced during streptozotocin-induced diabetes. [25] Thus measurement of hydroxyproline content is an index of collagen turnover. In present study there was a marked decrease in hydroxyproline levels in diabetic rats as compared to normal control. But treatment with *Lepidium sativum* orally as well as topically significantly increased the levels of hydroxyproline which further resulted in formation of granulation tissue over the wound. Hexosamine is a component of the ground substance required for the synthesis of the extracellular matrix and thus for proper wound healing. In general, the level of hexosamine increases between 7th–12th post wounding day and then decreases slowly. [26,27] We evaluated hexosamine content in granulation tissues of excision wounds in order to monitor the wound healing process. FA significantly increased hexosamine content in both orally and topically treated groups compared to the diabetic control rats indicating improved extracellular matrix synthesis and thus proper wound healing. In this study we observed that there was impairment in healing of wound in diabetic rats, also time taken and wound healing rate by diabetic rats was longer than time taken by normal rats. Treatment with *Lepidium sativum* orally as well as topically showed faster wound healing when compared to diabetic control rats. Among all groups the best wound healing rate was observed in higher oral dose (30 mg/kg) which was almost similar to standard group (mupirocin).The wound healing activity was evidenced by percentage wound closure was better by orally treated groups than topically treated.

CONCLUSION

This study evaluated the wound healing activity of the crude extract of the seeds of *Lepidium sativum linn.* The results indicated that the ethanolic extract of seeds increases the % wound closure, hydroxyproline and hexosamine levels and also significantly decreased the wound area, blood glucose and blood clotting time. Thus, the results of the present study suggest that

crude extracts of the plant can be used for treating the wounds and other dermatological disorders in diabetic condition.

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