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RESEARCH ARTICLE

Study of the Efficacy of Aloe Vera Extracts in Treatment of Non-Infected Wounds Induced by Sulferric Acid and Infected Wounds with *Staphylococcus aureus*.

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Abstract

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S. aureus poses an important problem in hospitals, nursing homes, and other health care settings. Serious infections due to these organisms currently necessitate the use of non- β -lactam antibacterial therapy. H₂SO₄ comes from strong poisons, because the symptom poison appears after five minutes from apple on skin. If possible treatment by Na₂Co₃ as antidot for H₂SO₄, but the necrosis or bacterial infection cases should by use drugs. *Aloe vera* gel is used topically for its anti-inflammatory and wound-healing properties, but it has also been used internally as a general tonic. The aim of this study is to evaluate the efficacy of aloe vera extract to treatment of normally induced and infected wounds with *Staphylococcus aureus*.

In this study Twenty rabbits were used in this experiment. All of these rabbits were males. Burning techniques: These animals were divided in four groups and burning induced by H₂SO₄. After 1 hour of washing with distilled water, group 1 was still without treatment, group 2 was treated with aloe Vera extract for 7 days, group 3 was infected with staphylococcus aurous and group 4 was infected with staphylococcus aureus and aloe Vera extract for 14 days. Re-isolation of bacteria: Swabs were taken from all experimented burns every 24 hours before and after treatment to detect the progress of infection and recovery respectively. The result appeared whilst signs of burns and infected animals were recorded after 24-48 hours of infection as redness, ulceration, edematous and thickening of skin tissue was shown in non infected group I. The extract used for group II of non infected animals showed some signs after nearly 2 days, however after 7 days the wound progressed to healing and there was no swelling and ulceration. The skin began to soften and there was no cracked tissue. Infected group III, with staphylococcus aureus, showed thickening areas and swelling with pus cell. Group IV was infected with staphylococcus aureus and treated with aloe Vera extract for 14 days. There was not any inflammation areas caused by bacteria pus cell and swelling and they showed a degree of healing. While Histopathological sections appeared in the skin of the rabbits at 14 days, post-wound of the extracts shows granulation tissue in the wound area, irregular fibrous connective tissue proliferation with congested blood vessel, as well as mononuclear cell infiltration in some areas and an edematous. These results in the present study prove that aloe Vera extracts have the ability to accelerate burn healing time and act as an antibacterial through inhibition. Staphylococcus aureus were experimentally infected in burns and therefore are recommended to be used for treatment of wounds and burns.

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INTRODUCTION

S. aureus poses an important problem in hospitals, nursing homes, and other health care settings. Serious infections due to these organisms currently necessitate the use of non- β -lactam antibacterial therapy [1]. Many hospitalacquired MRSA strains are only susceptible to vancomycin [2]. Thus, there are strong concerns about the possible development and spread of vancomycin resistance in MRSA. Some vancomycin-resistant MRSA strains have been reported since 1996 [3,4]. Some necrosis poisons cases occur by strong acids as H₂SO₄. It effects skin created necrosis or burns and these allow for bacteria growth. H_2SO_4 is one form of strong poison, because the poison's symptoms appear after five minutes from application on skin. If possible, treatment by Na₂Co₃ as antidote forH₂SO₄, but the necrosis or bacterial infection cases should use drugs. Aloe Vera gel is used topically for its antiinflammatory and wound-healing properties, but it has also been used internally as a general tonic. The main constituents of Aloe Vera gel are mucopolysaccharides (glucomannans, polymannoses, about 10% of total solids), enzymes, anthranoids, lignin, saponins, vitamins, amino acids (almost 50% of the total amount consisting of 8 of the 10 essential amino acids) and minerals (quantities not given). Total solids are in the range of 1.3 to 2%, the rest being water[5]. Aloe Vera gel is obtained either from hand-filleted leaves of Aloe barbadensis or, by cold processing of the whole leaf, in which case the product usually also contains appreciable quantities of the latex material and anthranoids. The anthranoids in whole leaf extracts of Aloe Vera can however be reduced to levels below10mg/kg in the product [6, 7, 8].

Oliver[9] indicates that *Aloe Vera* gel is used in veterinary medicine topically to promote wound healing on general skin wounds in all animals. It has also been recommended as a teat-dip in lactating cows, by intra mammary administration for (adjuvant) treatment of mastitis or high somatic cell counts, and by oral route in all food-producing species as adjuvant treatment for a number of afflictions (ranging from anemia to infertility, mastitis and shock[8,9]. Medicinal plants according to the World Health Organization (WHO) defines them as herbal preparations made by introducing plant materials to extraction, fractionation, purification, concentration, or other physical or biological processes, which may be produced as a basis for herbal products or for immediate consumption. Plants are rich in nutrients and they are the main source of food. Plants are also rich in compounds, which have pain relieving and healing abilities. In human medicine *Aloe Vera* gel is used topically to promote wound healing. Oral use as a general tonic for a number of indications, where scientific proof is outstanding, has also been described. *Aloe Vera* gel is also widely used in cosmetics [10,11,12,13]. Aloe Vera has ulcerogenic activity[36]

Materials and methods:

1. Plants extract :

The seeds of the *aloe vera* leaf were collected from local Medicinal plants in AL-Qassim city.

2. **Preparation of the extract:**

Aloe Vera leaf, 200g, was dissected into small portions and then put in a blender mix. The mixture was dissolved by 750 ml of 70% hydro alcoholic solution in a mechanical shaker (magnetic starrier) at 55°C for 5 hours. The content was filtered and kept in an incubator at 37°C for 36hours. The concentrated extract was stored dry at -20°C in deep freezers. The percentage yield of the parsley 2.4% weighting extract by electrical imbalance according to dose used in this study, re-extracted another amount of leaf so as to get enough quantity.

3. Experiment and grouping of animal:

A-Laboratory animals :Twenty rabbits were used in this experiment. All of these rabbits were males and their weight ranged between 1.5-2kg. Six month olds were obtained from the Laboratory Animal Colony, AL-Qassim University. The rabbits were kept under controlled hygienic conditions in wood cages and fed a basil diet for one week before starting the experiment for acclimatization.

B- Material: Four material were used in this experiment. They were aloe Vera extract, aloe Vera oil, Na2Co3 and H₂SO₄.

C-burning techniques: Rabbits were clipped and shaved around 5 cm diameter in the area of the lateral part of the body. Then animals were divided in four groups and burning was induced by H_2SO_4 . After 1 hour of washing with distilled water:

Group I was still without treatment.

Group II was treated with aloe Vera extract for 7 days.

Group III was infected with staphylococcus aureus.

Group IV was infected with staphylococcus aureus and treated with aloe Vera extract for 14 days.

D- **Re-** *isolation of bacteria*: Swabs were taken from all experimented burns every 24 hours before and after treatment to detect the progress of infection and recovery respectively.

Result:

1. Infected burns symptoms:-

The signs of experimental burns and infected animals were recorded after 24-48 hours of infection as redness, ulceration, edema and thickening of the skin tissue, as shown in non infected group I in picture (1). The extract used for group II of non infected animals showed some signs nearly after 2 days, however after 7 days the wound progressed to healing and there was no swelling and ulceration, skin began to soften and there was no cracked tissue, as shown in picture 2. Infected group III with staphylococcus aureus showed thickening area and swelling with pus cell and were treated with aloe Vera extract for 7 days, picture 3. Group IV was infected with staphylococcus aureus and treated with aloe Vera extract for 14 days. There is no inflammation area by bacteria pus cell and swelling and they showed a degree of healing, while non-treatment showed necrosis by infection of bacteria, as shown in picture 4.



Picture (1): group I not infected by burn after 24-48 hours



Picture (2): GroupII used to extract after 7 days.



Picture(3): Infected group III with *staphylococcus aureus* and used oil and oil with cream.



Picture (4): Group IX was infected with *staphylococcus aureus* and treated with cream and non treatment.

2. Re-isolation of infected bacteria:-

To detect the presence of bacteria in the infected burns, swabs from the burns were sub-cultured for three days (24,48,72 hours) before treatment on blood agar. The results in 72 hours are shown in photo (1).



Photos (1) bacterial growth before treatment.

3. Histopathological study:

This study found Histopathological section in the skin of normal rabbits at 7 days. Post-wound shows fibrous connective tissue proliferation and some areas and an edematous, as shown in picture 5.



Picture(5):Histopathological section in the skin of a normal rabbit at 7 days post-wound (H&E stain 40X).

While Histopathological section in the skin of rabbit at 7days post-wound induced by H_2SO_4 shows vessels and in some fibroblasts it shows irregular fibrous connective tissue proliferation with congested blood vessels, as well as mononuclear cell infiltration, as shown in picture 6.



Picture 6:Histopathological section in the skin of rabbit at 7 days post-wound induced by H_2SO_4 (H&E stain 40X).

On other hand, Histopathological section in the skin of rabbits at 14 days, post-wound, in the extract group shows densely arranged collagen fibers below the dermo-epidermial junction, in addition to fibroblasts, PMN's and macrophages, as shown in picture 7.



Picture 7: Histopathological section in the skin of a rabbit at 14 days post-wound in an extract group (H&E stain 40X).

While, Histopathological section in the skin of rabbits at 14 days post-wound of the extract shows granulation tissue in the wound area, irregular fibrous connective tissue proliferation with congested blood vessel, as well as mononuclear cell infiltration in some areas and an oedematous, as shown in picture 8.



Picture 8: Histopathological section in the skin of a rabbit at 14 days post-wound of extract (H&E stain 40X).

Discussion

The signs of experimental burns and infected animals were recorded after 24-48 hours of infection as redness, ulceration, edematous and thickening of skin tissue, as shown in the non infected; group I. The extract used for group II of non infected animals showed some signs after nearly 2 days, however after 7 days the wound progressed to heal and there was no swelling and ulceration, skin become softer and there was no cracked tissue. On the other

hand, the group of animals infected with bacteria and treated by aloe Vera extract showed degrees of healing. There was no pus cells or inflammation in the areas and the burn was healing completely after twenty days, unlike the non treated infected group where there was thickening areas with pus cells. The activity of A. Vera acts against both Gram-positive and Gram-negative bacteria has been indicated. Anthraquinones isolated from the exudates of A. Vera have shown wide antimicrobial activity. Many anthraquinones have shown antiviral and/or veridical effects on enveloped viruses[14,15] Extract of A. Vera showed significant in vitro antibacterial efficacy against Staphylococcus aureus. These results agree Obata [16], Agarry, et al[17], Thu[18] indicate that aloe Vera have antibacterial, antiviral and antifungal activity. Extracts of Aloe Vera is a proven skin healer. This may be due to contain sources of vitamins, minerals, antifungal, antiviral, antibacterial, more over astringent agent. Aloe Vera also has stimulatory effect on immune systems increasing antibody so to stimulate wound healing by means of growth factor, such as gibberellins, auxin and mannose phosphate. The Manos phosphate of *aloe Vera* activates insulin-like growth factor receptors that aid entry of glucose to a cell [19]. Aloe vera molecules can stimulate the fibroblast to increase collagen and proteoglycan. Protein parts of aloe vera act as a guide to the poly saccharide chain into the receptors to the mechanism of action of aloe vera pain reliever and anti-inflammatory [20]. Several mechanisms have been proposed for the wound healing effects of the aloe extract, which involve keeping the wound moist, accelerated epithelial cell migration, increased maturation of collagen and reduction in inflammation [21]. Choi et al [22] indicates that A. Vera contains A 5.5 kD a glycoprotein, which showed an increase in cell migration and accelerated wound healing in a human keratinocyte monolayer. In a raft culture, it exhibited stimulation of epidermal tissue formation as well as marked expression of proliferation markers on the immunohistochemical level. The enhanced wound healing effect and cell proliferation of this glycoprotein fraction was confirmed in hairless mice[23]. Aloe vera helped to soothe skin injuries affected by burning, skin irritations, cuts and insect bites, and its bactericidal properties relieved itching and skin swellings [24]. It is known to help slow down the appearance of wrinkles and actively repair the damaged skin cells that cause the visible signs of aging. Aloe is a powerful detoxifier, antiseptic and tonic for the nervous system [25]. It also has immune-boosting and anti-viral properties. Research has proven that adding aloe vera to one's diet improves digestion. As a general health tonic, Aloe vera is a useful source of vitamins. Aloe vera gel contains a large range of vitamins - including vitamin B12, Vitamin A, B-Group vitamins, Vitamin C, Vitamin E and folic acid [26,27]. Antioxidants protect the cell against reactive oxygen/ nitrogen species (ROS/RNS) by scavenging the free radicals in the cellular milieu. The increased levels of ROS/RNS generated during irritation and irradiation have been shown to be effectively scavenged by some antioxidants present in plants [28]. Moreover aloe Vera has antioxidant properties, reducing free radical-induced tissue injury [29]. Much research shows that aloe Vera extracts act as a cell proliferators, healer, demulcent and allergy reducer. Topically it is used for skin ulcers, burns, irritations and bites. (Rajesh et al [30], Retee et al.[3] indicate that aloe Vera gives excellent results after topical application on burns of first and second degree nature. However, it is believed that these biological activities should be assigned to a synergistic action of the compounds contained therein, rather than a single chemical substance, due to A. Vera gel increased the *in vitro* skin penetration. A. Vera gel could potentially be used as a penetration enhancement agent [32]. Hosseinimehr et al [33] indicates that rats treated with Aloe

Vera powdered gel 0.5% and silver sulfadiazine at 24 hours of burn injury, induced by hot water, show after 25 days of treatment that aloe Vera appears to decrease wound sizes to 0.78 cm2. silver sulfadiazine also shows aloe Vera treated burned skin biopsies showed significant wound healing by repithelialization of the epidermis and fibrosis of the dermis. The inflammation and granulation tissue formation was minimal and bacteria was not found in this specimen. On the other hand, much research on humans shows aloe vera gel treated partial thickness burns and the wound healed quicker (11.8 days) than the Vaseline gauze (18.2 days) and other topical medication taking a longer time[34,35]. *Aloe Vera* contains salicylic acid which is an aspirin-like compound with anti-inflammatory analgesic and anti-bacterial properties[37]. It has anti-pyretic properties for reducing fevers. Topical used aloe Vera for second degree burn in dog will accelerate the burn wound healing process and decrease degree of inflammation and exudation when comparison with both the control and Silver Sulfadiazine group [38]

Conclusion

Aloe vera extracts have the ability to treat burns induced in rabbits by sulfuric acid and act as an antibacterial through inhibition of staphylococcus *aureus experimentally infected in burns*. It also showed shorter wound care time for skin sights in animals who were treated with *aloe Vera*. Therefore it is recommended to be used by patients and animals in the treatment of itching, wounds, burns and in cosmetics.

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