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Full Length Research Paper

Comparative study of antimicrobial activities of *Aloe vera* extracts and antibiotics against isolates from skin infections

Asma Bashir[1], Bushra Saeed[1], Talat .Y. Mujahid[2] and Nayar Jehan[2]

[1]Stem Cell Research Center, SZABIST, Karachi, Pakistan.
[2]Department of Microbiology, University of Karachi, Karachi, Pakistan.

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Plants are of relevance to dermatology for both their beneficial and adverse effects on skin and skin disorders. One of the medicinal plants, *Aloe vera* (*Aloe barbadensis* Miller), is reputed to have medicinal properties. For centuries, it has been used for an array of ailments such as mild fever, wounds and burns, gastrointestinal disorders, diabetes, sexual vitality and fertility problems to cancer, immune modulation, AIDS and various skin diseases. In this study, antibacterial activity of leaf and gel extracts of *A. vera* were tested against gram positive and gram negative skin infections isolates. For this purpose, one hundred and fifteen bacterial strains were isolated from skin wounds, burns and acne patients from various hospitals of Karachi, a cosmopolitan and heavily populated city of Pakistan, and the strains were identified by conventional methods. Among the total isolates, 90% of the organisms were gram positive while the remaining 10% were gram negative. The gel extracts of *A. vera* showed antibacterial activity against both gram positive and gram negative isolates while the leaf extracts showed no such activity. In parallel, five standard antibiotics were also tested against the isolated strains. The data showed promising results in case of *A. vera* compared to five broad-spectrum antibiotics. Additionally, the study also demonstrated that the skin infectious isolates were resistant against broad-spectrum antibiotics.

Key words: *Aloe vera*, antimicrobial, antibiotics, skin infections, Karachi.

INTRODUCTION

Aloe vera (*Aloe barbadensis* miller) is a plant, which belongs to the family of Liliaceae and is mostly succulent with a whorl of elongated, pointed leaves (Strickland et al., 2004; Beckford and Badrie, 2000). The name is derived from the Arabic word 'alloeh' which means 'bitter', referring to the taste of the liquid contained in the leaves. The term ALOE refers to a solid residue obtained by evaporating the latex derived from the outer layers of the plant leaf. Taxonomists now refer to *Aloe barbadensis* as *Aloe vera*. The central bulk of the leaf contains colourless mucilaginous pulp, made up of large, thin walled mesophyll cells containing the *A. vera* gel itself. Despite its wide use as a folk remedy over a long period of time, the biochemical details of its action on physiological/ pathophysiological functions have not been systematically investigated (Rajasekaran et al., 2006; Tanweer et al., 1996). The plant has a long history as a multipurpose folk remedy, and has been associated with myth, magic and medicine since pre-biblical times. Historical evidence indicates that *A. vera* originated in the warm, dry climate of Southern and Eastern Africa, and was subsequently introduced into Northern Africa, the Arabian Peninsula, China, Gibraltar, the Mediterranean countries, and the West Indies (Jassim and Naji, 2003; Pribitkin and Boger, 2001). *A. vera* is described as one of the most talked about, yet most misunderstood plants in history. Modern clinical use of *A. vera* began in the 1920s and claims now abound, in numerous research and commercial literature in journals and on the Internet, regarding its numerous therapeutic potentials when used both topically and parenterally. It is acclaimed to cure ailments ranging from mild fever, wounds and burns, gastrointestinal

*Corresponding author. E-mail: asmibashir@gmail.com.

disorders, diabetes, sexual vitality and fertility problems to cancer, immune modulation and AIDS (Taiwo et al., 2005; Grindlay et al., 1986; Mackay and Miller, 2003).

Medicinal plants of the lily family (Liliaceae), genus *Aloe*, have been used for the treatment of skin diseases for more than 5000 years. Among more than 360 *Aloe* species, *A. vera* (*A. barbadensis* miller) has been the most popular in both folk and officinal medicine (Larry, 2003; Sun et al., 2002; Kodym and Bujak, 2002). *A. vera* extracts are widely used in a variety of over-the-counter and dermatological products. Many studies report the effective use of this plant when applied topically for the treatment of burns, sunburns, inflammatory skin disorders and wounds (Belo et al., 2006; Reider et al., 2005; Paulsen et al., 2005). *A. vera* is a plant that can produce latex and gel. The gel is extracted from the leaf, and it is this substance that is most used as a treatment. *A. vera* has been evaluated in a number of different clinical contexts and some promising results have been found for its use in controlling cardiovascular risk factors and diabetes, besides being beneficial in areas of dermatology. One explanatory factor for this is the anti-inflammatory properties of the plant (Davis et al., 2006; Choi et al., 2001; Tian et al., 2003). It contains over 70 biologically active compounds and is claimed to have anti-inflammatory, anti-oxidant, immune boosting, anti-cancer, healing, anti-ageing and anti-diabetic properties. *Aloes*, by contrast, is an anthraquinone derivative of the sap of the *Aloe* leaf which has been used for centuries as a purgative (Langmead et al., 2004; Gallagher and Gray, 2003). *A. vera* gel has been widely promoted and used by patients for the treatment of a range of inflammatory digestive and skin diseases (Langmead et al., 2004; Lee et al., 2004). The antibacterial activities of *A. vera* were dependent on the dose of anthraquinone. It is reported that *A. vera* possesses antifungal, antiviral, antibacterial and acaricidal activity against skin infections such as acne, herpes and scabies (Mantle et al., 2001; Hart et al., 1990). It contains a compound that neutralizes and binds with FGF-2 receptor, or otherwise alters signaling pathways for FGF-2 by affecting both GJIC and proliferation of diabetic fibroblast (Abdullah et al., 2003). Several reports suggest that beneficial effects of *Aloe* gel are due to its high molecular weight components such as polysaccharide, lectin like proteins and prostaglandins (Kodym et al., 2003; Puke and Ayensu, 1985; Koo, 1994).

The aim of the present study was to evaluate the effects of an *A. vera* gel and leaf extract on skin infection isolates. The results obtained with *A. vera* were compared with five different standard antibiotics.

MATERIAL AND METHODS

Collection of samples

Skin infection isolates were obtained from septic wounds and burns patients undergoing injury dressing at different hospitals. Wound exudates were obtained from the infected sites of each patient with sterile cotton swabs and applied to freshly prepared slants of nutrient agar and Mannitol Salt agar (Oxoid). The cultures were then transferred to the laboratory where they were incubated at 37°C for 24 h (Kolawole and Shittu, 1997; Huys et al., 2002).

Bacterial isolates, culture media and species identification

Colonies growing on slants were streaked on top of freshly prepared plates of Mannitol Salt agar and Brain Heart Infusion agar and incubated again. Primary characterization of isolates was based on the Gram stain, morphological and cultural characteristics. Identification also includes growth on different media including Nutrient agar and Brain Heart Infusion agar, fermentation on Mannitol Salt agar (Oxoid). Catalase and coagulase tests were also performed for biochemical characterization (Udo et al., 2006).

Maintenance of clinical isolates

Stock cultures were maintained in vials by growing the skin isolates in 3 ml nutrient broth and next day overlaying with 3 ml 40% glycerol. Vials were than frozen at -70°C (Gul et al., 2004; Richardson et al., 2005).

Determination of antibiotic resistance profile

Skin isolates were subjected to antibiotic resistance screening by disk diffusion method. For this purpose inocula were prepared by diluting overnight cultures in sterile sodium chloride (0.9%) suspension and then match with the 0.5 standard Mac Farland index. Bacterial suspensions were then plated onto Mueller-Hinton agar (Oxoid) and the commercially available antibiotic discs were placed on lawn of culture and the plates were incubated over night at 37°C (Hoeger, 2004; Veronica and Keelan, 2006). Sensitivity, intermediate sensitivity, and resistances were determined by the zone of complete growth inhibition around each disk according to reference standards. The following antibiotic discs were used: methicillin (10 ug), bacitracin (15 ug), vancomycin (30 ug), novobiocin (30 ug) and erythromycin (15 ug).

Aloe vera gel and extracts

A. vera plants were purchased from a nursery in Karachi. The gel was taken from the leaves into a clean container and used as such (Agarry et al., 2005). While the leaves from which the gel has been drained were air dried (50 g), macerated with 100 ml sterile distilled water in a warning blender for 10 min. The macerate was first filtered through doubled layered muslin cloth, then centrifuged at 4000 g for 30 min.The supernatant fluid was filtered through Wattman No.1 filter paper and heat sterilized. The extract was preserved aseptically in a brown bottle at 5°C until used (Satish et al., 1999; Sayaka and Watanabe, 2003).

Antimicrobial susceptibility testing of *A. vera*

Sterile agar (at 45°C) was poured into sterile Petri dishes, which had been inoculated with the test organisms. The plates were allowed to gel for an hour. Wells (10 mm diameter) were made with the aid of flamed cork borer on the surface of the agar plates. About 0.1 ml of each of the gel and the leaf extracts were delivered into each of the wells. These were incubated at 37°C for 24 h. The presence of zones of inhibition was regarded as the presence of

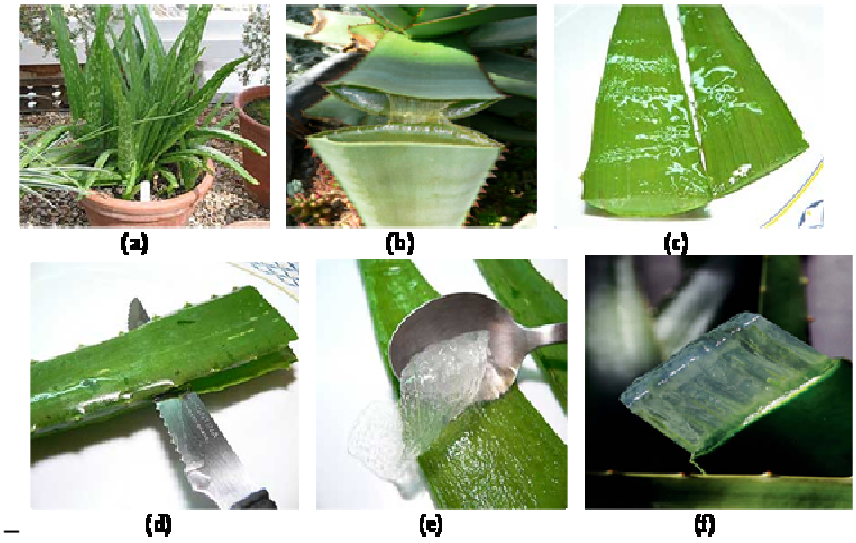

Figure 1. *Aloe vera* plant. (a) Succulent, dense, clumps of fleshy, light green leaves. (b) Incision in the leaf. (c) Leaf unwraps from the upper side. (d) Leaf unwraps from both sides (e) Removal of *A. vera* gel from the leaf. (f) Section of *A. vera* gel.

Table 1. Percentage of gram positive and gram-negative clinical skin infection isolates.

Isolate	Total number of organism	Total percentage
Gram Positive	**100**	
Staphylococcus aureus	55	47.8
Staphylococcus epidermidis	35	30.4
Streptococcus pyogenes	10	8.6
Gram Negative	**15**	
Pseudomonas aeruginosa	15	13.0

antimicrobial action. From the inhibition zones seen, antimicrobial activity was expressed in terms of average diameter of the zones of inhibition measured (Agarry et al., 2005).

RESULTS AND DISCUSSION

A. vera is described as one of the most talked as regards, yet most misunderstood plants in olden times and the *A. vera* plant has been used for an array of ailments, including skin diseases (Figure 1). Numerous studies report the effectual use of this plant when functioning topically for the healing of burns, sunburns, inflammatory skin disorders and wounds (Belo et al., 2006; Reider et al., 2005; Paulsen et al., 2005). The present study was designed to identify the antibacterial activity of A. vera leaf and gel which was also compared with five standard antibiotics against clinical isolates from community-acquired skin infections. The antibacterial activity was monitored using agar-well diffusion and agar disc diffusion method; activity was determined by noting the zones of inhibition around the wells or discs (Gul et al., 2004). For this purpose, one hundred and fifteen clinical isolates of skin infections from different clinical laboratories of Karachi were isolated and identified by conventional methods. Antibacterial activity of *A. vera* leaf and gel was checked against these isolates. Table 1 showed that the percentage of gram-positive isolates was as follows: *Staphylococcus aureus* (47.8%), *Staphylococcus epidermidis* (30.4%) and *Streptococcus pyogenes* (8.6%) while the percentage of gram negative isolates includes: *Pseudomonas aeruginosa* (13.0%). *A. vera* has been used as a cosmetic and medical remedy since ancient times and has gained increasing popularity in recent years. Despite its widespread use, reports of allergic reactions are rare (Reider et al., 2005).

A. vera is used in cosmetics, drinks, detergents, as well as in stockings, diet foods, toothpaste and clothing. It is marketed as a remedy for atherosclerosis, allergies, AIDS, prevention of radiation-induced dermatitis, wound healing, psoriasis, insomnia, cancer, pulpitis and several other diseases. Scientific studies investigating these claims are few in number, and the majority of them have

Table 2. Comparative study of *A. vera* leaf and gel with standard antibiotics against gram positive and gram-negative clinical skin infection isolates.

Antibiotic	Gram positive (%)	Gram negative (%)
Methicillin	68.0	60.8
Bacitracin	25.0	51.4
Vancomycin	80.5	72.2
Novobiocin	54.1	63.6
Erythromycin	55.6	42.4
A. vera Leaf	0.0	0.0
A. vera Gel	75.3	100.0

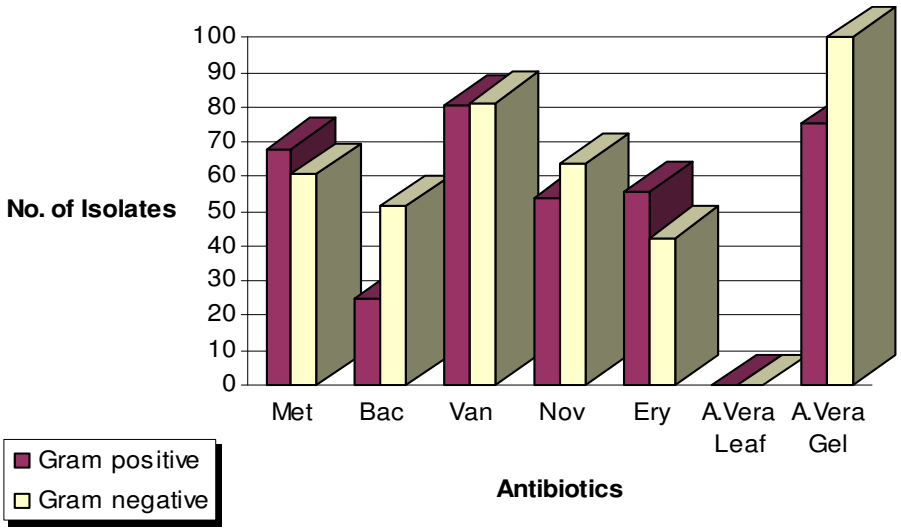

Figure 2. Comparative study of *A. vera* leaf and gel with standard antibiotics against clinical skin infection isolates.

been unable to diminish the intuitive scepticism against miracle cures, like *A. vera* seems to be (Reider et al., 2005). Table 2 showed the comparative study of *A. vera* leaf and gel with standard antibiotics. The results showedthat *A. vera* leaf was 0% effective against the entire tested gram positive as well as gram-negative isolates. *A. vera* gel showed 100% activity against gram-negative isolates and 75.3% against all tested gram-positive isolates. This result could be responsible for the popular use of *A. vera* gel and leaf to relieve many types of gastrointestinal irritations (Foster, 1999; Grindlay and Reynolds, 1986), since *S. aureus* form part of the normal microbial flora of the skin, upper respiratory tract and intestinal tract (Cheesbrough, 1984). Also the gel is also said to promote wound healing due to the presence of some components like anthraquinones and hormones (Davis, 1997), which posses antibacterial antifungal and antiviral activities. However, most of the constituents are found in the gel and not in the leaf, hence the gel is likely to be more active than the leaf. The gel possesses 100% inhibitory effect on *P. aeruginosa* while the leaf had no effect. *P. aeruginosa* is known to cause skin infection especially at burns sites, wounds, pressure sores and ulcers. Traditionally attributed to the limited permeability of the *P. aeruginosa* outer membrane, it is now clear that the organism's intrinsic multidrug resistance also owes much to the operation of broadly specific antimicrobial efflux systems (Poole and Srikumar, 2001). The inhibitory effect of the gel of *A. vera* on the growth of *P. aeruginosa* gives an explanation of its reputation as a healing plant for burns.

In our studies, the most effective antibiotic for gram positive is vancomycin showing 80.5% efficacy, then methicillin with 68.0% efficacy, erythromycin with 55.6% efficacy, novobiocin with 54.1% efficacy and bacitracin with 25.0% efficacy. The most effective antibiotic for gram negative is vancomycin showing 72.2% efficacy, then novobiocin with 63.6% efficacy, methicillin with 60.8% efficacy, bacitracin with 51.4% efficacy and erythromycin with 42.4% efficacy. The susceptibility of gram positive isolates to erythromycin, methicillin and vancomycin was generally high while that to bacitracin and novobiocin was low. This suggests that the penicillinase-resistant anti-bacterial agents should be selected as a first choice to

treat these infections (Hoeger, 2004). The comparative antimicrobial activities are graphically represented in (Figure 2). The mechanisms by which *A. vera* gel may act are unclear. *In vivo*, *A. vera* reduces irritant-induced pro-production of inflammatory mediators in paw, ear and synovial models of inflammation in animals. *A. vera* gel and its components also ameliorate ultravioletinduced immune suppression. *In vitro*, several fractions of *A. vera*, as well as the unfractionated whole gel, have anti-oxidant effects. *A. vera* gel contains peroxidase activity, 28 several super oxide dismutase enzymes 29 and a phenolic antioxidant (Langmead et al., 2004). Davis (1997), in his experiment challenged the medical views of the relationship between AIDS and HIV infections and *A. vera*. He sees a promising role for this natural brood spectrum healing plant because of its immunodulatory properties and can also act as an immune stimulant. In a multicenter investigation, patch testing to *A. vera* yielded no evidence of a sensitizing potential for this herbal drug. This is remarkable in view of the fact that a large number of the tested subjects had sought medical help because of dermatitis due to other causes and were obviously at risk for skin reactions. None of them reported any side-effects after the application of *A. vera* on the skin. It may be assumed that an even much larger number of the patients had experienced unwitting contact to *Aloe*-containing products in the past (Choi et al., 2001).

Conclusion

The present study has revealed the importance of natural products to control antibiotic resistant bacteria, which have been a threat to human health. Numerous plants indigenous to Pakistan in general have been found with amazing medicinal properties. Some are well-evaluated vis-à-vis their content of specific active principles against the target microorganism while others are not. It is therefore highly essential that medicinal plants whose properties have not been fully characterized should form a top agenda of top management in developing nations whose citizens are sometimes unable to afford expensive orthodox medicine. This policy, if pursued, will not only preserve the scarce foreign exchange but also promote the spirit of plant conservation.

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