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Full Length Research Paper

A comparative analysis of two medicinal plants used to treat common skin conditions in South Africa

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Infectious dermatological diseases are a common occurrence in southern Africa. Plants showing dermatological properties are highly sought after due to their ability to stop bleeding, speed up wound healing and to soothe skin exposed to burns (Lewis and Elvin-Lewis, 1977). An attempt was made to validate the use of *Haworthia limifolia* and *Aloe excelsa* against microbial properties from extracts of leaves against five Gram positive, four Gram negative bacteria and six species of fungi. All Gram positive bacteria were inhibited by both the ethyl acetate and acetone extracts for leaves of *H. limifolia*. However, only one strain of Gram negative bacteria was inhibited by the same extracts. Ethyl acetate extract of *A. excelsa* was only effective against three Gram positive bacteria whilst acetone extracts were effective against all bacteria except for *Shigella sonnei* and *Enterobacter aerogene*. Both ethanol and aqueous extracts of *H. limifolia* and *A. excelsa* showed antifungal activity. *H. limifolia* extracts showed greater antibacterial activity than *A. excelsa* whilst *A. excelsa* showed greater antifungal activity than *H. limifolia*. Use of either species as traditional medicine will therefore depend on the type of infection or condition presented by the patient.

Key words: Haworthia limifolia, Aloe excelsa, antimicrobial, traditional medicines.

INTRODUCTION

Infectious dermatological diseases are a common occurrence in rural parts of South Africa. According to the World Health Organization (WHO) more than 80% of the world's population relies on traditional medicine for their primary health care needs. Traditional healing plays an integral part of black African culture with the majority of people consulting traditional healers (Lindsey et al., 1999). Mander (1998) conservatively estimated that at least 50% of the population in KwaZulu-Natal is rural and that the frequency of indigenous medicine use is likely to be greater than the urban areas given the limited access to western bio-medical services in remote areas.

South Africa has a very rich plant biodiversity, many of which are medicinally useful (Afolayan and Adebola, 2004). This rich resource is decreasing at an alarming rate as a result of indiscriminate and unsustainable harvesting. Demand for plant derived medicines has

*Corresponding author. E-mail: rogercoopoosamy@gmail.com. Tel: +27 82 200 3342. Fax: +27 31 907 7665. created a trade in indigenous plants in South Africa conservatively estimated to be worth approximately R270 million a year (Mander, 1998). Mander (1997) estimates that there are 27 million consumers of traditional medicine and its supporting industry in South Africa. Population growth coupled with rapid urbanization is creating an ever increasing demand for traditional medicine.

Plants showing dermatological properties are highly sought after due to their ability to stop bleeding, speed up wound healing and to soothe skin exposed to burns (Lewis and Elvin-Lewis, 1977). Although skin diseases do not usually threaten life, their unforgiving itching can cause misery and their presence may be a social stigma. In sub-Saharan Africa skin conditions are dominated by bacterial and fungal infection and their clinical expression is often modified by HIV-induced immune- suppression (Naafs, 2004). Skin diseases may therefore constitute a large percentage of all attendees in clinics in rural communities in South Africa. Despite the plethora of antibiotics afforded by lower plants such as fungi, microbial diseases are still on the rise in developing countries due

Table 1. Traditional uses of Aloe exc	əlsa.
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Plant Part	Uses	Preparation Used
Leaves	Skin burns	Leaf exudates
Leaves	Rashes	Diluted tea from leaf exudates.
Leaves	Sun burns	Diluted tea from leaf exudates.
Leaves	Laxatives	Leaf exudates
Leaves	Blood purifers	Leaf exudates
Leaves	Skin irritants	Leaf exudates
Leaves	Moisturizers	Diluted tea from leaf exudates
Leaves	Immune boosters	Diluted tea from leaf exudates

to the relative unavailability of medicines and the emergence of wide scale drug resistance (Okeke et al., 2005). The fleshy leaves and roots of most species within the Aloe family are used in many traditional treatments (Mabberley, 1987). Traditional healers and indigenous people utilize mainly the leaf sap of this genus widely for the treatment of wounds, burns, rashes, cracked lips, and cracked skin (Cera et al., 1980). Another much used species, exhibiting similar properties, is *Haworthia limifolia*. In this study we attempted to confirm the antimicrobial activity of crude extracts of *H. limifolia* and *A. excelsa* currently being used for the treatment of skin diseases. Both these species are widely available in multi markets although increased use as resulted in pressure on wild populations, thus, escalating trade prices.

MATERIALS AND METHODS

All plant material was collected and a voucher specimen was stored as Cooper03/2009 (Aloe excelsa) and Cooper02/2010 (*H. limifolia*) at Mangosuthu University of Technology. Specimen verification was done with the help of Plant Specialists at Ezemvelo KZN Wildlife.

Antibacterial assay

Methods used in extraction of plant extract by traditional healers was performed according to Coopoosamy et al. (2010). Briefly, according to Coopoosamy et al. (2010), leaf material of *H. limifolia* and *A. excelsa* were collected from the field and dried in an oven at 60°C until sufficiently dried (4 days). One kilogram of dried material for each specimen was then crushed and placed in a 2 L conical flask containing one of three mediums that is water, ethyl acetate and acetone, for extraction. The media except the boiled medium (method used by traditional healers) were left for 72 h in an orbital shaker at 20 shakes per minute. After 72 h the extracts were filtered. The boiled medium was placed on a hot plate at constant 80^0 c for a period of 4 hours after which it was filtered. The extracts were then used for further tests.

The plant extracts were then tested for antibacterial properties against five strains of Gram-positive (*Bacillus subtilis, Micrococcus kristinae, Bacillus cereus, Staphylococcus aereus, Staphylococcus epidermidis*) and four strains of Gram-negative bacteria (*Escherichia coli, Proteus vulgaris, Enterobacter aerogenes,* and *Shigella sonnei*) for antibacterial activity. Each organism was prepared by diluting in 24 h old broth cultures with sterile nutrient broth. The cultures were then diluted 100 fold to give approximately 10^{6} bacteria ml⁻¹.

Antifungal assay

H. limifolia and A. excelsa leaves (approximately 1 kg) were cut into small pieces and crushed in a homogenizer. Different extraction methods were use to determine the most suitable for extraction of compounds for testing against microbials. Each method would result in different compounds being extracted as per the polarity of the solvents. The plant materials were soaked in ethanol (95% v/v) and in distilled water in 2 L conical flasks for 3 weeks. The extracts (water and ethanol) obtained were evaporated at reduced pressure (45°C) to a residue. Extracts for testing ethanol and aqueous extracts were prepared in three different concentrations. The stock solutions were prepared by dissolving 100 mg of dry extract in 1 ml of ethanol and water separately in order to obtain a concentration of 100 mg/ml dilutions (1:10, 1:100, 1: 500). These stock solutions were then used in phosphate buffer at pH 6.0 to evaluate the antifungal activity (Champion et al., 1992). The solutions were then tested for antifungal activity using the following fungal cultures: Aspergillus flavus, Aspergillus glaucus, Candida albicans, Candida tropicalis, Trichophyton mentagrophytes, and Trichophyton rubrum. Plates containing potato dextrose agar were used to serve as controls.

RESULTS AND DISCUSSION

In Southern Africa as well as in China and Mexico, the leaf gel or exudates of *Aloe* sp. are used in various dermatological remedies, such as, minor skin irritations (Grindley and Reynolds, 1986). Although used as medicinal remedies for centuries, the only three common uses that predominate are the laxative effect, blood purifying effects as well as external treatment of skin infections or injuries (Table 1).

The traditional use of *H. limifolia* involves use both in spiritual as well as traditional medicinal practices (Table 2). The traditional use has been linked to sores, burns and sun-burns. Skin diseases occur in various forms, basically classified as non-contagious diseases, the primary of which are bacterial, fungal, viral and parasitic diseases. These diseases occur throughout the world, but are prevalent in the rural and tropical regions (Davies

Table 2. Traditional uses of H. limifolia.

Plant Part	Uses	Preparation Used
Whole Plant	Removal of evil	Plant used as a charm
Leaves	Blood purifiers	Diluted tea from leaf exudates.
Leaves	promote pregnancy	Diluted tea from leaf exudates.
Leaves	Treatment of sores	Leaf exudates
Leaves	Treatment of superficial burns	Leaf exudates
Leaves	Treatment of sun burns	Leaf exudates
Leaves	Cleansing of digestive system	Diluted tea from leaf exudates

Table 3. Minimal inhibitory concentration (MIC) of *H. limifolia* antibacterial assay on crude extract (Controls:Chloramphenicol and Streptomycin sulfate) n = 3.

Bacteria	Grom 1	Medi	Control (µg/ml)			
(10 ⁶ Bacteria/ml)	Gram +/-	Cold water	Ethyl acetate	Acetone	Chlor ^a	Strept
Bacillus subtilis	+	Na	3.0	3.0	<2.0	<2.0
Micrococcus kristinae	+	Na	4.0	5.0	<2.0	<2.0
Bacillus cereus	+	Na	4.0	4.0	<2.0	<2.0
Staphylococcus aureus	+	Na	4.0	5.0	<2.0	<2.0
Staphylococcus. epidermis	+	Na	5.0	5.0	<2.0	<2.0
Escherichia coli	-	Na	5.0	5.0	<2.0	<2.0
Proteus vulgaris	-	Na	Na	Na	<2.0	<2.0
Shigella sonnei	-	Na	Na	Na	<2.0	<5.0
Enterobacter aerogene	-	Na	Na	Na	<2.0	<2.0

Na = No activity, All tests were done in triplicates and the averages are indicated, $Chlor^{a} = Chloramphenicol$, Strept ^b = Streptomycin sulphate.

et al., 1986).

All gram positive bacteria were inhibited by both the ethyl acetate and acetone extracts of leaves of H. limifolia (Table 3). However, only one strain of gram negative bacteria was inhibited by the same extracts. In contrast, there were no inhibitory effects on Gram positive and negative bacteria with aqueous extracts (Table 3). Gram positive bacteria often cause human diseases such as colds, wounds and sores (Waihenva et al., 2002), Medically relevant Gram negative cocci cause sexually transmitted diseases. meningitis and respiratory whilst bacilli symptoms species primarily cause respiratory, urinary and gastrointestinal problems. It is therefore evident that leaf extracts from H. limifolia may play a valuable role in treating various skin conditions caused by Gram positive bacteria. Traditional healers tend to boil large quantities of leaf material of H. limifolia, thereby possibly releasing the bioactive compounds that are involved in the treatment process.

Ethyl acetate extracts of *A. excels*a were only effective against three Gram positive bacteria whilst acetone extracts were effective against all bacteria except for *Shigella sonnei* and *Enterobacter aerogene* (Table 4). The ability of *Aloe* to inhibit growth of micro-organisms has been demonstrated by rapid clearing of infected tissue after induction of therapy. In a study of *Aloe* in the treatment of tuberculosis during the early stages, it was found that moderate antibacterial activity was exhibited from the leaf sap (Droscoll et al., 1974; Ghannam et al., 1986).

Both ethanol and aqueous extracts of H. limifolia and A. excelsa showed antifungal activity (Table 5). The antifungal activity of the ethanol was shown to be slightly more effective than the aqueous extracts with the 1:10 dilution producing the maximum rates of inhibition. Growth inhibition (zone of inhibition) was recorded as very high (++++), high (+++), medium (++), and low (+), which indicated zones of inhibition between 41 to 50, 31 to 40, 21 to 30, and 11 to 20 mm, respectively (Coopoosamy, et al., 2010). It was evident that extracts from A. excelsa possessed greater antifungal activity across the dilution range (Table 5). It is therefore not surprising that there has been a recent explosion of Aloe products on the market, mostly cream based, which is used frequently to treat dry skin, sun burns, itching, irritated skin, rashes, scabies etc.

Skin fungus infections include itching, redness and thickened skin of fungal infections can look like other burn occurs, the patient loses the protective epithelial layer of skin and as a result is prone to infection by

Bacteria	Crow 1	Med	Control (µg/ml)			
(10 ⁶ Bacteria/ml)	Gram +/-	Cold water	Ethyl acetate	Acetone	Chlor ^a	Strept
Bacillus subtilis	+	Na	3.0	2.0	<2.0	<2.0
Micrococcus kristinae	+	Na	4.0	1.0	<0.2	<2.0
Bacillus cereus	+	Na	2.0	2.0	<2.0	<2.0
Staphylococcus aureus	+	Na	Na	1.0	<2.0	<2.0
Staphylococcus. epidermis	+	Na	Na	1.0	<2.0	<2.0
Escherichia coli	-	Na	Na	3.0	<2.0	<2.0
Proteus vulgaris	-	Na	Na	2.0	<2.0	<2.0
Shigella sonnei	-	Na	Na	Na	<2.0	<5.0
Enterobacter aerogene	-	Na	Na	Na	<2.0	<2.0

Table 4. Minimal inhibitory concentration (MIC) of A. excelsa antibacterial assay on crude extract (Controls:Chloramphenicol and Streptomycin sulfate) n = 3.

Na = No activity, All tests were done in triplicates and the averages are indicated, $Chlor^{a}$ = Chloramphenicol, Strept ^b = Streptomycin sulphate.

Table 5. Effect of ethanol and aqueous extracts obtained from H. limifolia and A. excelsa on different fungal specimens.

	H. limifolia						A. excelsa					
Fungal species	Ethanol extract			Aqueous extract		Ethanol extract			Aqueous extract			
	1:10	1:100	1:500	1:10	1:100	1:500	1:10	1:100	1:500	1:10	1:100	1:500
Aspergillus flavus	+++	++	+	+	+	-	++++	+++	++	++++	+++	++
Aspergillus glaucus	+++	+	+	+	+	-	++++	+++	+++	+++	+++	+++
Candida albicans	+++	++	+	+	-	-	++	+	-	++	+	-
Candida tropicalis	+++	++	+	+	+	-	+++	++	++	+++	++	+
Trichophyton mentagrophytes	++	+	+	+	-	-	+++	++	++	++	++	+
Trichophyton rubrum	++	+	+	-	-	-	++	++	+	++	+	+

- = Negative antifungal activity, + = Positive antifungal activity (low inhibition), ++ = Positive antifungal activity (medium inhibition), +++ = Positive antifungal activity (high inhibition), ++++ = Positive antifungal activity (very high inhibition). N.B.: Plates containing Potato dextrose agar only served as controls. Control did not show any inhibition of any of the test fungal species.

Table 6. Minimal inhibitory concentration observed in different concentrations prepared from stock solution of 100 mg/ml of aqueous and ethanol extracts of *H. limifolia* and *A.excelsa* (n = 3).

-	H. lii	nifolia	A. excelsa			
Fungal species	Ethanol extract	Aqueous extract	Ethanol extract	Aqueous extract		
Aspergillus flavus	1:500	1:100	1:500	1:500		
Aspergillus glaucus	1:500	1:100	1:500	1:500		
Candida albicans	1:500	1:10	1:100	1:100		
Candida tropicalis	1:500	1:100	1:500	1:500		
Trichophyton mentagrophytes	1:500	1:10	1:500	1:500		
Trichophyton rubrum	1:500	1:10	1:500	1:500		

opportunistic infection such as *C. albicans. C albicans* is readily introduced into burn wounds because this yeast is commonly a member of the normal flora found on mucous membranes. Anecdotal evidence suggests that when *Aloe* is used on patient's burns, the frequency of fungal infections decrease (Lee et al., 1999). The minimum inhibitions observed are given in Table 6.

Conclusion

It was clearly evident from the study that both H. limifolia

and *A. excelsa* possess antibacterial and antifungal properties. However, *H. limifolia* extracts showed greater antibacterial activity than *A. excelsa* whilst *A. excelsa* showed greater antifungal activity than *H. limifolia*. Use of either species as traditional medicine will therefore depend on the type of infection or condition presented by the patient.

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