

African Journal of Infectious Diseases Research ISSN 2756-3340 Vol. 8 (8), pp. 001-004, August, 2022. Available online at www.internationalscholarsjournals.org © International Scholars Journals

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Full Length Research Paper

The use of *Lawsonia inermis* linn. (henna) in the management of burn wound infections

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Accepted 6 July, 2022

The effects of water and chloroform extracts of the leaves of *Lawsonia inermis* (henna plant) against the primary invaders of burnt wounds was investigated. Clinical isolates of *Staphylococcus aureus, Streptococcus* sp, *Pseudemonas aeruginosa, Candida albicans, Fusarium oxysporum*, and *Aspergillus niger* were treated with extracts of the leaves of *L. inermis* for antimicrobial activity using *in vitro* agar incorporation method and well diffusion methods respectively. The henna leaves extracts were able to inhibit the growth pattern of *A. niger* and *F. oxysporum*. *Streptococcus* sp. and *S. aureus* were also inhibited by the extracts. Inhibition of the microorganisms' growth suggests that henna may be valuable in the management of burnt wound infections.

Key words: Lawsonia inermis, burnt wounds, henna.

INTRODUCTION

Many of today's modern drugs have their origin in traditional plant medicine (Blanks et al., 1998). The therapeutic efficacies of many indigenous plants for several disorders have been described by practitioners of traditional herbal medicines (Natarajan et al., 2003). Natural products are a significant source of synthetic and traditional herbal medicine and are still the primary health care system (Singh and Singh, 2001). Being sources of many life sustaining metabolites, the research is still on for plants to be used in healing. This in part is due to the growing problem of worldwide antibacterial resistance.

Isolation of microbial agents less susceptible to regular antibiotics and recovery of resistant isolates during antibacterial therapy is increasing throughout the world (Bonjar, 2004). One of the measures of combat this increasing rate of resistance is to have continuous investigations into new, safe and effective antimicrobials as alternative agents to substitute with less effective ones. Because of this, research is being carried out to investigate ethnobotanical uses of plants prevailing among native people. The henna plant is one such plant known since with healing attributes, and is now the subject of intense scientific study.

The henna plant is a glabrous much branched shrub or quite a small tree with grayish-brown bark. Leaves are opposite, sub-sessile, elliptic or broadly lanceolate, entire, acute or obtuse, 2-3 cm long and 1-2 cm wide. Flowers are numerous, small white or rose coloured and fragrant. The plant constituent are made up of mannite, tannic acid, mucilage and gallic acid, but the main constituent is 2-hydroxynapthoquinone (lawsone), known to be the major bioactive constituent.

Lawsone is the chief constituent responsible for the dying properties of the plant. Dried powdered leaves of henna contain about 0.5-1.5% lawsone, traditionally used to produce colorfast orange, red and brown dyes. Henna is native to a number of tropical regions in Asia, Northern Africa and Australia. It is naturalized and cultivated in the tropics of America, Egypt, India and parts of the middle east.

Complication due to burn wounds arise from the colonization of the burn site by such organisms as *Streptococci* sp., *Straphylococcus aureus*, *Pseudomonas aeruginosa*, *Fusarium oxosporum*, *Aspergillus niger* and *Candida alblicans*. These complications can usually be

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Test Isolate	Extract	Concentration	Zone of inhibition (mm, diameter)			
C. albicans	Water	10 mg/ml	0			
		30 mg/ml	0			
		60 mg/ml	0			
		80 mg/ml	0			
	Chloroform	10 mg/ml	0			
		30 mg/ml	0			
		60 mg/ml	0			
		80 mg/ml	0			
Streptococcus	Water	10 mg/ml	0			
		30 mg/ml	9			
		60 mg/ml	15			
		80 mg/ml	23			
	Chloroform	10 mg/ml	0			
		30 mg/ml	11			
		60 mg/ml	18			
		80 mg/ml	24			
S. aureus	Water	10 mg/ml	11			
		30 mg/ml	18			
		60 mg/ml	20			
		80 mg/ml	25			
	Chloroform	10 mg/ml	10			
		30 mg/ml	17			
		60 mg/ml	21			
		80 mg/ml	26			
P. aeruginosa	Water	10 mg/ml	0			
		30 mg/ml	0			
		60 mg/ml	0			
		80 mg/ml	6			
	Chloroform	10 mg/ml	0			
		30 mg/ml	0			
		60 mg/ml	0			
		80 mg/ml	8			

Table 1. Effects of chloroform and water extracts of the leaves of *L. inermis* against the yeast *C. albicans* and the bacteria *Streptococcus* sp., *S. aureus*, and *P. aeruginosa*.

avoided by good initial therapy. Penicillin therapy is usually undertaken in hospital. However, in third world countries such as Nigeria where medical care is sometimes substandard, patients stay home and nurse themselves back to health using local remedies. Some common practices are the application of tea leaves, aloe vera gel in its crudest form, castor oil, egg yolks or the dried leaves of henna plants. The effect however of the use of these materials leaves much to the imagination. Since rural people have developed methods in taking care of their burn wound by the application of a paste made from the dried and powered leaves of the henna plant, there is a need to study the significance of this plant in the treatment of burn wound infections.

MATERIALS AND METHODS

Plant samples were obtained from the Botanical Garden of the Usman Danfodiyo University Sokoto, Nigeria and identified at the herbarium of the Botany Unit. Fresh leaves were collected and dried in the oven at a temperature of 60°C. The dried leaves were ground to a powder.

Extract preparation

400 g of the powder was suspended in 500 ml of distilled water in one litre flask for 24 h at room temperature. The mixture was then sieved through a fine muslin cloth followed by filtration using filter paper to trap the finer particles that went through the cloth. The filtrate was then mixed with the chloroform in a separating funnel, the mixture shaken until separation was observed in form of two

Test Isolates	Extracts	Concentrations	Growth at different days (mm)						
			1	2	3	4	5	6	
Control			8.3	29.1	38	57	81.7	90	
A. niger	Water	10 mg/ml	8.0	28	35	56	80	90	
		30 mg/ml	5.3	26	31.5	53	79.1	90	
		60 mg/ml	4.5	20	25	46	61	74.5	
		80 mg/ml	0	0	0	7.5	14	20	
	Chloroform	10 mg/ml	7.5	25	30	52	71	88	
		30 mg/ml	5	23	29	50	70	78	
		60 mg/ml	4	18	23	41	57	70.5	
		80 mg/ml	0	0	0	0	0	0	
F.oxysporum	Water	10 mg/ml	6.1	13	2.5	4.5	66	79	
		30 mg/ml	0	0	6.1	9.3	15.2	19	
		60 mg/ml	0	0	0	5	10.3	17.5	
		80 mg/ml	0	0	0	0	0	0	
	Chloroform	10 mg/ml	6.8	15	27	47.1	72	85	
		30 mg/ml	0	0	5.2	10.3	17.3	23	
		60 mg/ml	0	0	0	41.1	9.5	16.4	
		80 mg/ml	0	0	0	0	00		

Table 2. Effects of water and chloroform extracts of the leaves of L. inermis against the mold A. niger and F. oxysporum.

layers; the water and the chloroform extract. The different layers were run out into separate beakers and placed in an oven to dry at 50°C. Residues of the extracts were made into suspensions using sterile distilled water at concentrations of 10, 30, 60 and 80 mg/ml.

Specimen cultures

Bacterial and fungal cultures used in this study were obtained from clinical isolates obtained at the Usman Danfodio University Teaching Hospital Sokoto, Nigeria from burn wound patients. Bacterial pathogens included in the study were *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Streptococcus* sp. The fungal pathogens used include *Aspergillus niger*, *Fusarium oxysporum* and *Candida albicans*.

Inoculum preparation

The bacterial pathogens were grown on nutrient agar and maintained on slants while the fungi were grown on potato dextrose agar. A cork borer with dimensions 2×2 was used to make impressions in the culture containing the fungi so the size of the inoculum placed at the centre of each test and control plate was 2×2 for the fungi except *C. albicans* with many bacteria-like features which was treated like the bacteria.

Determination of diameter of growth inhibition

For the fungi, agar incorporation method was used (*C. albicans* excluded). Five (5 ml) of each concentration of the extracts (chloroform and water) was measured out and mixed with 15 ml of melted media and poured into sterile petri dishes and left to solidify. The plates were inoculated with each of the various fungi. Suitable controls were included for each. The plates in duplicate for each agent were incubated at room temperature for 6 days being the amount of time it took for the control plate to fill up. The plates were

observed on a daily basis and readings taken for each day using the linear measurement method and the mean was recorded.

For the bacteria and *C. albicans*, the agar well diffusion method was employed. Four 9 mm ditches or wells were made in each plate of solidified nutrient agar. Five (5 ml) of each concentration of the extracts was measured out and mixed with 0.3 g of agar. The well was carefully filled with this mixture and left to solidify. Each of the plates were seeded with each of the different organisms and incubated in the incubator for 24 h. The measurements were then taken again using linear measurement method.

RESULTS AND DISCUSSION

Antibacterial activity is recorded when the zone of inhibition is greater than 6 mm. The implication here being that for S. aureus, antibacterial activity was reported for all concentrations in both the water and chloroform extracts (Table 1). For P. aeruginosa, only mild antibacterial activity was recorded at the highest concentration in both the water and chloroform extracts. In Streptococcus, there was no activity at the lowest concentration but as the concentration increased antibacterial activity was recorded. Extracts had no activity against the yeast C. albicans. For the fungi, A. niger and Fusarium, there was a slight inhibition in growth (Table 2). Still as the concentration increased, inhibition was accomplished for the first 2-3 days and eventually there was complete inhibition evident in the chloroform extracts.

The present study showed that henna leaves extracts were capable of inhibiting the growth of microorganisms that are involved in causing burn wound infections. This finding therefore support the use of henna in the management of burn wound infection. Use of henna in management of burns may offset the complication that arise in the use of conventional wound dressings such as silver nitrate which imparts stains and is time consuming apart from being able to cause hyponatraemia or hypokalaemia as well as the use of mafenide (sulphamylin) which can be painful thus distressing the patient.

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