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Full Length Research Paper

Bioactivity Profiling of *Hydnora abyssinica* A. Braun Against Diverse Microbial Groups: Fungi and Bacteria

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Root samples of *Hydnora abyssinica* were collected from Wad Medani city, central Sudan, to examine their antimicrobial potentials. Root extracts, aqueous, methanolic and chloroform, were obtained and bioassayed *in vitro* for ability to inhibit growth of 6 human pathogenic fungi and 4 types of bacteria, the differences in bioactivity of aqueous, $CHCL_3$ and MeOH extracts were analyzed. Despite extreme fluctuations in activity, the undiluted aqueous extract was the most inhibitory followed by methanolic extract, while the chloroform extract was the least effective. Growth of pathogens was inhibited by all extracts in a concentration dependent manner. Phytochemical analyses showed preponderance of condensed tannins and phenols in the plant root extracts. The results confirmed the antibacterial and antifungal activity of the roots and support the traditional use of the plant in healing bacterial infections, the possibility of therapeutic use of Sudanese *H. abyssinica* as antimicrobial agent is worthy of further investigations.

Key words: *Hydnora abyssinica*, medicinal plants, antibacterial activity, antifungal activity, Sudan.

INTRODUCTION

Some bacteria and fungi are extremely pathogenic causing serious human infections. The discovery of antibiotics to combat these pathogens marked a revolution in the 20th century (Evan, 1992). However, because of inappro-priate use of antibiotics in human and veterinary medi-cine, certain strains of bacteria and fungi developed the ability to produce substances which block the action of antibiotics and/or change their target or ability to pene-trate cells (Groove and Randall, 1955). Therefore, disease-causing microbes that have become resistant to antibiotic drug therapy are an increasing public health problem. Tuberculosis, gonorrhea, malaria and childhood ear infections are just a few of the diseases that have be-come hard to treat with antibiotics (Misra, 1977; Goun et al., 2003). Development of resistance to synthetic drugs has revived interest in antibiotics of botanical origin.

Plants have been used to treat human, animals and plant diseases from time immemorial. Many of today's modern and effective drugs have their origin in herbal traditional folk medicine (Misra, 1977; Goun et al., 2003; Natarajan et al., 2003). *Hydnora abyssinica* (A. Braun), a root holoparasitic plant on several *Acacia sp.* in central Sudan, is renowned for healing attributes and has been used by practitioners of traditional medicine to cure severe bacterial infections (Almaqbool et al., 1985; Almaqbool et al., 1988). The plant belongs to the family *Hydnoraceae* which is predominant in the semiarid regions of Africa and in the southern Arabian peninsula (Musselman and Visser, 1989; Musselman, 1991). More than 12 *Hydnora* species have been described and their antimicrobial properties were thoroughly investigated (Bolin et al., 2009).

Paucity of pharmacological and chemical data on *H. abyssinica* (Sudanese tartous) prompted the present investigation on its antimicrobial activities and efficacy against different pathogenic fungal and bacterial species.

MATERIALS AND METHODS

Plant material

The study was undertaken at the department of microbiology and molecular biology, university of Al -Neelain, Khartoum, Sudan, during August to December 2008. *H. abyssinica* (A. Braun) root samples were collected from a round *Acacia sp.* at Wad Medani city central Sudan. The plant was identified and authenticated by Dr.

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Table 1. Comparative bioactivity of *H. abyssinica A*. Braun against different fungal species*.

Type of extract	Concentration (mg/ml)	Fungal species					
		Ef	Ma	Tr	Tc	T t	Ca
Water	10	16.08 ± 0.12	15.11 ± 0.55	15.24 ± 0.05	16.00 ± 0.22	16.11± 0.52	16.18 ± 0.78
	50	17.12 ± 1.03	16.32 ± 1.13	17.77 ± 1.15	17.18 ± 1.25	17.12 ± 1.05	16.15 ± 0.28
	100	17.14 ± 0.42	17.22 ± 0.03	18.28 ± 1.65	17.95 ± 0.35	17.25 ± 0.85	17.22 ± 0.48
Methanol	10	15.15 ± 1.05	13.21 ± 0.65	12.7 ± 0.45	12.95 ± 1.05	14.15 ± 1.00	16.12 ± 0.11
	50	16.21 ± 0.75	14.77 ± 0.15	13.30 ± 1.98	13.33 ± 1.08	14.23 ± 0.08	16.11 ± 0.77
	100	16.15 ± 0.55	15.61 ± 1.16	14.13 ± 1.16	14.00 ± 1.99	14.22 ± 0.99	15.13 ± 0.44
Chloroform	10	14.72 ± 0.65	13.52 ± 0.35	13.33 ± 0.75	13.10 ± 0.25	15.03 ± 0.65	15.11 ± 0.55
	50	16.25 ± 1.15	14.14 ± 1.22	13.13 ± 0.98	15.66 ± 0.98	15.11 ± 0.27	15.28 ± 0.88
	100	16.15 ± 0.55	16.30 ± 1.00	14.11 ± 1.44	16.14 ± 1.04	15.21 ± 0.14	15.22 ± 0.66
Nystatin (reference drug)	25	18.83 ± 0.85	18.25 ± 0.41	18.22 ± 0.76	19.25 ± 0.61	18.52 ± 0.41	18.20 ± 0.01

*Data are presented as mean ± SD of zone of inhibition (mm); inhibition zones are the mean of 5 replicates; *Ef = E. floccosum, Ma = M. audouinii, Tr = T. rubrum* , *Tc = T. concentricum, T t = T. tonsurans*, *Ca = C. albicans*.

Alwaia A.Al-Awad of biology department, university of Al- Neelain, Khartoum, Sudan. Voucher specimens were deposited at the biology department Herbarium.

Preparation of extracts

Dried roots were ground to powder. Samples (10 g each) of the coarsely powdered roots were successively Soxhlet extracted with $CHCl_3$ and MeOH for 24 h. The extracts were evaporated under vacuum and the residues were separately dissolved or suspended in the same extracting solvent (10 ml) and kept in a refrigerator till used. In addition, water extracts were prepared by adding distilled water to 10 g of coarsely powdered plant material in a conical flask and left to soak overnight at room temperature. The residue was then filtered under suction through filter paper and the final volume was adjusted to 10 ml with distilled water and the filtrate was imme-diately used.

Fungal species

6 fungal species, isolated from clinical cases, were provided by Khartoum educational hospital, Sudan. The fungi were *Epidermophyton floccosum, Microsporum audouinii, Trichophyton rubrum*, *Trichophyton concentricum, Trichophyton tonsurans* and *Candida albicans*. Each of the fungi was cultured on Sabouraud's dextrose agar medium (Oxoid) and incubated at 25°C for 7 days, to obtain inocula for testing.

Bacterial strains

4 types of bacteria namely *Staphyllococcus aureus, Bacillus sub-tilis, Escherichia coli* and *Pseudomonas aeruginosa* were used. They were isolated from clinical cases and provided by Khartoum educational hospital, Sudan. The bacteria were cultured on nutrient broth (Oxoid) at 37°C for 24 h.

Determination of antifungal activity

Sterile, filter paper discs of 6 mm diameter were impregnated with about 0.1ml/disc of extract which have been dissolved in dimethyl sulphoxide (DMSO) and placed in duplicates onto the Sabouraud's dextrose agar plates, seeded with 0.2 ml of fungal suspension. The plates were then incubated at 25°C for 10 - 14 days (Ugarte et al., 1987; Iqbal Zafar et al., 2002). The zone of inhibition around each disc was measured in mm. The results are presented as means ± SD. Nystatin (25 µg/disc) was used as a reference standard drug for comparison. To determine the influence of tartous extracts on the growth of 6 different fungal dermatophytes, aliquots of different dilutions of the extracts (10, 50 and 100 mg/ml) were separately added in 175 ml Czapek-Dox liquid medium in conical flask. Flasks containing Czapek-Dox medium alone (200 ml) were included as controls for comparison. The flasks, inoculated with *E. floccosum, M. audouinii, T. rubrum*, *T. concentricum, T. tonsurans*, and *C. albicans* were incubated at 25°C for 15 days. Each set of treatments was replicated 5 times. The fungal mats, harvested through Whatman filter paper No. 2, pressed between blotting paper to remove excess were weighed to determine mycelial fresh weight.

Antibacterial activity

Each root extract (water, methanol and chloroform) was tested against the four types of bacteria using the cup-plate agar diffusion method (Groove and Randall, 1955) and the inhibition zones were measured. Means diameters of the inhibition zones were reported in Table 3.

Phytochemical screening

Phytochemical screening was conducted for root samples using the method of Crombie et al. (1990) and preliminary phytochemical analysis results of *H. abyssinica* roots were reported.

RESULTS AND DISCUSSION

As a general rule, plant roots are considered active against both fungi and bacteria when the zone of inhibition is greater than 6 mm (Evans, 1992). All extracts of *H. abyssinica* roots, irrespective of concentration, significantly ($p = 0.05$) suppressed growth of the tested organisms (Tables 1 and 3). The aqueous extract was clearly superior in bioactivity as compared to those of methanol and chloroform. The maximum inhibition zone

Table 2. Influence of *H. abyssinica* A. Braun roots on the growth of different fungal dermatophytes*.

Fungal species	Mycelial fresh weight (g) grown on Czapek-Dox medium					
	L.S.D. (at 5 %)	D.S.D. (at 1%)	100 mg/ml	50 mg/ml	10 mg/ml	0 (control)
Epidermophyton floccosum	0.653	1.111	8.59	9.66	10.11	16.11
Microsporum audouinii	1.973	1.777	7.65	8.23	9.67	18.33
Trichophyton rubrum	1.126	1.763	9.11	9.34	11.45	17.11
Trichophyton concentricum	1.542	1.983	10.55	10.98	10.15	15.22
Trichophyton tonsuran	1.231	2.194	7.66	9.23	11.44	14.33
Candida albicans	0.111	1.529	7.88	8.66	8.71	13.12
C.D. at 1%			2.111	1.222	1.600	1.809
C.D. at 5%			1.321	1.770	1.346	1.333

*Values are means of 5 replicates.

Table 3. In vitro antibacterial activity of H. abyssinica A. Braun roots*.

Type of extract	Zone of inhibition(mm)			
	P.a	E.c	S.a	B.s
Water	10a	8a	6a	8b
Methanol	8b	9b	8b	6a
Chloroform	7b	6c	5c	6a
Ampicillin 40 ug/ ml	8	16	14	12

was found in 100 mg/ml water extract concentration and it was 18.28 mm in the fungal isolate of the pathogen *T. rubrum*, while the least inhibition zone in the same concentration was 17.14 mm for the pathogen *E. floccosum*. The activity increased with increasing extract concentration. Even in the lowest concentration of water extract, that is, 10 mg/ml, all of the fungal isolates showed substantial inhibition in their respective growth. The minimum inhibition zone in 10 mg/ml concentration was observed in *T. rubrum* with 15.24 mm inhibition zone (Table 1) as compared to 16.18 mm in *C. albicans* at the same concentration (Table 1) . The maximum inhibitory effect for methanol extract at 10 mg/ml was observed also against *C. albicans* where the zone of inhibition was 16.12 mm against standard antibiotic (18.20 mm) and a minimum activity was observed against *T. rubrum* (12.77 mm).The chloroform extract of the plant showed maximum activity against the *C. albicans* where the zone of inhibition was 15.11 mm compared to that of the reference drug (18.20 mm) and the weakest activity was observed against *T. concentricum* (13.10 mm) in the same concentration (10 mg/ml). Results also showed that the growth in culture media of the different clinical fungal species was suppressed when the water root extract of the plant was used in different concentrations (Table 2). % reduction in mycelial weight of the different tested fun-gal species was directly correlated to the concentration of the root extract (Correlation coefficient) (Table 2). The maximum reduction in mycelial weight (3.00 g) was observed in 100 mg/ml water extract concentration in *M. audouinii* and *T. tonsurans* followed by *C. albicans* (7.88 g) and *E. floccosum* (8.59 g).The minimum reduction, on the other hand, was brought about by the water extract concentration 100 mg/ml for the pathogen *T. concentricum* (10.55 g). However, the reduction in mycelial weight is decreased when the concentration of the extract was increased with no significant differences between the 3 used water extracts concentrations of each treatment. Even in 10 mg/ml as lowest concentration the reduction in mycelial weight of the different tested fungi was sig-nificant over controls (Table 2).

Results obtained from *in vitro* antibacterial activity showed that the 3 types of root extracts have substantial inhibitory effects against the 4 tested bacterial species (Table 3). Still the water extract was superior in suppressing the bacterial growth, followed by methanol and chloroform extracts. The maximum inhibition zone was observed against *P. aeruginosa* (10 mm) followed by *B. subtilis* (8 mm) and 6 mm for *S. aureus*. The weakest activity was observed in chloroform extract with a maximum zone of inhibition 7 mm observed against *P. aeruginosa* and a minimum zone of inhibition 5 mm in *S. aureus* (Table 3). This data is in close agreement with previous reports elsewhere using the same plant (Bonjar, 2004; Leemann, 1933).

Furthermore, the roots of the plant were phytochemically screened and the results are shown in Table 4. The roots of the plant showed the presence of tannins and phenols in high levels concentration. Other consti-tuents such as proonthocyanins were found in a mode-rate concentration while flavonoids were found in low concentration. However, the significance of this finding remains the area of further investigations as far as the chemical constituents of this plant are concerned. Also, it is not possible to make a direct correlation between the observed activity of the plant extracts *in vitro* and the actual effects when used *in vivo* for the diseases obser-ved by the indigenous people and traditional healers. Therefore, it is important that the plant should also be investigated to evaluate the significance of these ex-tracts, clinical role and the medical system of indigenous

Table 4. Preliminary phytochemical analysis of *H. abyssinica* A. Braun.

Constituent	Level*
Phenols	+++
Tannins	+++
Proonthocyanins	++
Flavonoids	+
Mucilage	+
Sterols and/or triterpenes	±
Glycosides	-
Saponins	-

*:+ = low concentration, ++ = medium concentration, +++ = high concentration, = ± traces, - = not detectable.

indigenous people. Further research is necessary to isolate and characterize their active compounds for pharmacological testing. The present study identifies Sudanese *H. abyssinica* as potential source of biological antimi-crobial, since it showed a high activity against wide spectrum of bacteria and fungi which enables only human pathogenic fungi and bacteria to be killed without any side effects and/or bacterial resistance as current syn-thetic antibiotics are doing and this specificity appears as additional point in the natural antibiotics research.

REFERENCES

Almaqbool AZ, Bashir AK, Farouk A, Salih AKM (1985). Antimicrobial Activity of certain Sudanese plants used in Folkloric Medicine; Screening for Antimicrobial Activity (IV). Fitoterapia 56:331-334.

Almaqbool AZ, Bashir AK, Salih AKM (1988). Antimicrobial Activity of certain Sudanese plants used in Folkloric Medicine; Screening for Antimicrobial Activity (V). Fitoterapia 69:59-62.

Azaizeh H, Fulder S, Said K, Khalil O (2003). Ethno-Botanical know-ledge of local Arab practitioners in the Meddle Eastern region. Fitoterapia 74:98-108.

Bolin JFEM, Musselman LJ (2009). Pollination Biology of *Hydnora africana* Thunb. (Hydnoraceae) in Namibia: Brood Site Mimicry with Insect Imprisonment. Int. J Plant Sci., 170(2): 157–163.

Bonjar SGH (2004). Screening for Antibacterial properties of some Iranian plants against two strains of *Eschericia coli.* Asian J. Plant Sci. 3(3): 310-314.

Crombie L, Crombie WML, Whiting DA(1990). Alkaloids of Khat (*Catha edulis*). Alkaloids 39:139-164.

Evan CW (1992). Trease and Evans Pharmacognosy, 13th Edition. Bailliere Tindall, London, pp. 758-762.

Goun E, Cunningham G, Chu D, Nguyen C, Miles D(2003). Antibacterial and antifungal activity of Indonesian ethnomedical plants. Fitoterapia 74(6): 592-596.

Groove DC, Randall WA (1955). Assay methods of Antibiotics. Medical Encyclopoedia,Third Edition,New York. pp. 24-55.

Iqbal Z, Mussarat S, Farrakh H, Sheraz B, Mohammad I, Shahida Z, Bashir A (2002). Antifungal properties of some indigenous plants from Peshawar Valley. Asian J. Plant Sci. 1(6):708-709.

Khattak SG, Gilani SN, Ikram M (1985). Antipyretic Studies on some indigenous Pakistani Medicinal Plants. J. Ethno -Pharmacol., 14:45-51.

Kujit J (1977). Haustoria of phanerogamic parasites. Ann. Rev. Phytopathol. 17: 91-119.

Leemann AC (1933). The anatomical and physiological relation between *Hydnora solmsiana* Dinter and its host. Anna. Bot. 47: 159-162.

Misra SK, Sahu KC (1977).Screening of some indigenous plants for antifungal activity against dermatophytes.Indian J. pharmacol. 9(4): 269-272.

Mitscher LA, Leu RP, Bathala MS, Wu WN, Beal JL (1972). Antimicrobial agents from higher plants.1.Introduction, rationale and methodology. Lloydia 35:157-166.

Musselman LJ (1991). The genus *Hydnora* (Hydnoraceae). *In* Ransom J K, Musselman LJ, Worsham AD, Parker C, editors Proceedings of the fifth international symposium on parasitic weeds, CIMMYT, Nairobi, Kenya. pp. 247-250.

Musselman LJ, Visser JH (1987). *Hydnora johannis* in southern Africa. Dinteria 19: 77-82.

Musselman LJ, Visser JH (1989). Taxonomy and natural history of *Hydnora* (Hydnoracecae). Aliso 12: 317-321.

Natarajan V, Venugopal PV, Menon T(2003). Effect of *Azadarichta indica* (neem) on the growth pattern of dermatophtyes. Indian J. Med. Microbiol. 21: 98-101.

Ugarte R, Quilhor W, Diaz B, Vera A, Fieder D(1987). Atlvdade antimicrobiana *in vitro* del acido epiforelico L. Acta Farm. Bonaerense. 6: 65-69.