Pakistan J. Sci. Ind. Res., Vol. 21, Nos. 5-6, October - December 1978

STUDIES ON THE RATIONALE OF AFRICAN TRADITIONAL MEDICINE
Part III. Preliminary Screening of Medicinal Plants for Antifungal Activity

A.N. Sawhney*

Department of Microbiology and Immunology, P.O. Box 20693, Faculty of Medicine, University of Dar es Salaam, Tanzania

M.R. Khan, G. Ndaalio, M.H.H. Nkunya and H. Wevers

Department of Chemistry, P.O. Box 35061, University of Dar es Salaam, Tanzania

(Received March 9, 1978)

Seven of the 18 (39 %) plants traditionally used as remedies for skin diseases in East Africa were found to inhibit the growth of *Trichophyton mentagrophytes* and *Candida albicans in vitro. Harrisonia abyssinica* and *Solanum incanum* were most active plants in this category. High level of *in vitro* antifungal activity was observed in some of the medicinal plants used for treatment of other miscellaneous diseases, e.g. *Emilia sagittata, Bonamia mossambiscensis, Securinega virosa, Sida serratifolia* and *Citrus aurantifolia.* The antibacterial and antimycotic activity of the plant extracts tested did not exhibit any clear-cut interrelationship. Most of the extracts having high levels of activity against *T. mentagrophytes* and *C. albicans* had no activity against *Staphylococcus aureus.*

INTRODUCTION

Of all the microbial infections of human beings, the diseases caused by fungi are the most difficult to modify in their course or to prevent and it is increasingly evident that the incidence of such diseases is mounting [1]. These infections both deep and superficial have a world-wide distribution but some of the clinical manifestations such as ringworm tend to be more severe and prevalent in moist tropics [2]. Perusal of the literature [3,4] and personal discussions with practitioners of African traditional medicine [5] in Tanzania (East Africa) revealed that a number of herbs are empirically used for treating skin diseases and many of them are claimed to be very effective. For example the fruit of *Solanum incanum,* a wide-spread weed in Tanzania and Kenya is extensively used for the treatment of cutaneous mycotic infections and other pathological conditions. According to a recent report [6] the therapeutic activity of the berries of *Solanum incanum* is attributed to their contents of solanine and related glycoalkaloids. Except for the above, the authors are not aware of any reported attempt at testing the plants included in the present investigation for *in vitro* antimycotic activity. The present study, involving *in vitro* screening of commonly used medicinal plants, forms a part of the general investigation on the rationale of African traditional medicine, and places on record some of the observations made.

*To whom correspondence should be addressed.

MATERIALS AND METHODS

A total of 124 plants were screened for activity against the common dermatophyte *Trichophyton mentagrophytes,* as well as *Candida albicans.* Of these, 18 plants classified as group A were those commonly used to treat skin diseases, while the remaining 106 plants, placed in group B were used as remedies for other miscellaneous ailments. The details on their nomenclature, parts of the plant used and the traditional uses of the active plants are given in the Table 1. The plants which showed no activity are also listed.

Plant Extract. The crude plant extract was obtained by extracting fresh powdered leaves, bark, flowers or whole plant (500g) with methylated spirit (1l) for 10 hr or until the extract was clear. The extract was then filtered through cotton-wool and the solvent removed under reduced pressure on a rotary evaporator using a water-bath.

Test Cultures. Trichophyton mentagrophytes was a 10-day old culture maintained on Sabouraud's dextrose-agar medium in the Department of Microbiology and Immunology, Faculty of Medicine, University of Dar es Salaam. The dermatophyte was isolated locally from a case of Athlete's foot. *Candida albicans* culture used in the present study was an isolate from a case of oral, thrush, maintained in the above Department.

Culture Medium. Sabouraud's dextrose-agar medium

(Oxoid) was used throughout the present investigations.

Bioassay Procedures. For preliminary screening of antifungal activity, 0.1 ml of the crude plant extract was incorporated in 3.0 ml of Sabouraud's dextrose-agar medium contained in culture bottles. The media containing the extracts were then inoculated with *T. mentagrophytes*, keeping adequate controls. The inoculated bottles were kept at room temperature and observed for growth of fungus up to a period of 6 weeks. The antifungal activity of an extract was assessed on the degree of inhibition of growth of the fungus in the media. Some of the extracts which exhibited high inhibitory effect on the growth of the fungus were also investigated for minimum inhibitory concentration. The extracts were further examined by filter-paper disc bioassay method by the procedures used by Beaman-Mbaya and Mohammed [6], and depending upon the diameter of the zone of inhibition, the antifungal activity was expressed as +, ++, +++, ++++ using the standards described elswhere [7]. In the case of *Candida albicans*, procedures already described for tests with *T. mentagrophytes* were followed, except that nystatin 100 mg discs (Mast Laboratories, Liverpool, England) were included as controls in filter-paper disc bioassay of antifungal activity.

RESULTS AND DISCUSSION

Among the 124 plant extracts tested, 33 (27%) showed *in vitro* antifungal activity. Of the 18 group A plant extracts 7 (39 %) showed activity ranging from + to +++ (Table 1) while 26 (25 %) of group B plants also exhibited similar *in vitro* inhibitory activity on both *T. mentagrophytes* and *Candida albicans* (Figs. 1 and 2).

A high level of antifungal activity (++++) was exhibited by extracts of *Emilia sagittata* DC., *Securrinega virosa* (pulp), *Sida serratifolia* (roots) and *Citrus aurantifolia* (roots). It is surprising to note that none of these plants are used to treat extremely prevalent dermatomycoses in East Africa. Instead these plants are used for miscellaneous ailments such as eye inflammation, topical dressing for wounds and contusions, diarrhoea, gonorrhoea, pneumonia, pulmonary tuberculosis and dysentery. Except for *Sida serratifolia* none of the above plants had any antibacterial activity *in vitro* when tested against *Staphylococcus aureus* (Oxford strain) and *Escherichia coli*. *Sida serratifolia* traditionally used as a remedy for gonorrhoea had significant antibacterial activity against *Staphylococcus aureus* (Oxford) but had no effect on the growth of *E. coli* [7] Among the 6 extracts having +++ *in vitro* antifungal activity 3 were also active against *Staphylococcus aureus* and 1 against *Escherichia coli*. A high degree of positive correlation between antifungal and antibacterial activity was observed in extracts with ++ antimycotic activity (73 %), while it was only 36 % in the case of extracts with + antifungal activity.

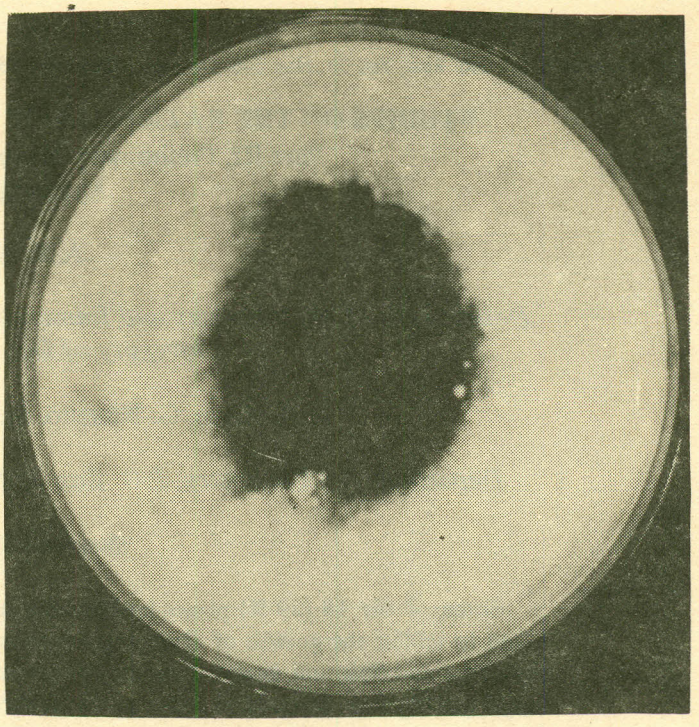

Fig. 1.

Fig. 2.

Zones of growth in hibition of *T. mentagrophytes*. (Fig. 1) and *Candida albicans* (Fig. 2) by *Citrus aurantifolia*. The standard NY100 is also given in Fig. 2.

Among the traditional remedies of skin diseases the highest *in vitro* antifungal activity was observed in the root bark extract of *Harrisonia abyssinica* followed by *Solanum*

Table 1. Susceptibility of fungus to various plant extracts.

Plant	Family	Part used and extracted	Traditional uses	Antifungal activity
Group A				
Plumeria rubra L.	Apocynaceae	Bark	Itching, diarrhoea, gonorrhoea, dropsy, purgative, skin diseases, warts, syphilis	++
Zizyphus pubescens Oliv.	Rhamnaceae	Leaves	Pneumonia, diarrhoea, dysentery, wounds, skin diseases	++
Solanum incanum L.	Solanaceae	Plant	Pneumonia, ringworms, liver disease, gonorrhoea, syphilis, earache	++
Solanum incanum L.	Solanaceae	Fruits	Dandruff, skin diseases, sores & wounds	++
Harrisonia abyssinica Oliv.	Simaroubaceae	Rootbark and twig	Skin diseases, haemorrhoids	+++
Waltheria indica L.	Sterculiaceae	Flowers	Skin diseases, syphilis, cleansing wounds, coughs, sores	+
Vitex fischeri Guerke.	Verbenaceae	Leaves	Chronic venereal diseases, epilepsy, as sedative, skin diseases	+
Group B				
Dictyophleba lucida (K. Schum.) Pierre.	Apocynaceae	Leaves		+++
Dictyophleba lucida (K. Schum.) Pierre.	Apocynaceae	Trunk		+++
Holarrhena febrifuga Klotzsch.	Apocynaceae	Leaves	Snake bite, venereal diseases, anti-dysentery	++
Ceiba pentandra Gaertn.	Bombacaceae	Leaves	Gonorrhoea and as dressings for wounds	+
Boscia salicifolia Oliv.	Capparidaceae	Bark	Rectal infections	++
Combretum zeyheri Sond.	Combretaceae	Whole plant	Diarrhoea	+++
Emilia sagittata DC.	Compositae	Whole plant	For inflammation of eyes, contusion, ulcerative processes, nasal disease, syphilis	++++
Bonamia mossambicensis (Klotzsch.) Hall. f.	Convolvulaceae	Leaves	Wounds	++++
Bonamia messambicensis (Klotzsch.) Hall. f.	Convolvulaceae	Roots	Wounds	+++
Bridelia cathartica B.	Euphorbiaceae	Stem	Purgative, stomach ache	+
Phyllanthus reticulatus P.	Euphorbiaceae	Plant	Gonorrhoea, ulcers, jaundice sores, uro-genital diseases	+
Pseudolachnostylis maprouneaefolia Pax.	Euphorbiaceae	Bark	Stomach ache, cathartic	++
Securinega virosa. (Willd.) Baill.	Euphorbiaceae	Pulp	Diarrhoea, gonorrhoea, pneumonia	++++
Cassia amiculata L.	Leguminosae	Bark	Headache, toothache	++
Xeroderris stuhlmannii T.	Leguminosae	Plant	Colds, chest troubles, elephantiasis	+
Asparagus falcatus L.	Liliaceae	Leaves	Syphilis	+
Hibiscus micranthus L.	Malvaceae	Plant	Earache, bronchitis, renal remedy	++
Sida serratifolia L.	Malvaceae	Leaves	Pulmonary tuberculosis, diarrhoea	+++
Sida serratifolia L.	Malvaceae	Roots	Gonorrhoea	++++
Citrus aurantifolia Swingle.	Rutaceae	Roots	Gonorrhoea, dysentery	++++
Fagara chalybea Engl.	Rutaceae	Rootbark	Diarrhoea, coughs, malaria, toothache	++
Deinbollia borbonica R.	Sapindaceae	Roots	Chest troubles, abdominal pains	+

Table 1 continued

Grewia forbesii Harv. ex. Mast.	Tiliaceae	Bark and roots	Rheumatism, lumbago and stiff neck	++
Premna chrysoclada G.	Verbenaceae	Leaves	Ulcers, venereal diseases	+
Cissus integrifolia Planch.	Vitaceae	Stem		+
Rhoicissus rovoilli Planch.	Vitaceae	Roots	Wounds, ophthalmic remedy	+

Plant extracts which did not show any *in vitro* antifungal activity: Acanthaceae: *Barleria prionitis* L. (roots, leaves and bark); Amaranthaceae: *Achyranthes aspera* L. (plant); Anacardiaceae: *Rhus natalensis* Bernh. (leaves), *Lannea stuhlmannii* Engl. (leaves); Annonaceae: *Anona senegalensis* Pers. (bark), *Uvaria acuminata* Oliv. (leaves), (roots); Apocynaceae: Calotropis gigantea Ait. F. (leaves), *Nerium oleander* L. (leaves); Araceae: *Stylochiton hennigii* Engl. (roots and leaves); Bignoniaceae: *Kigelia africana* Benth. (bark), *Tecomaria capensis* Spach. (leaves); Boraginaceae: *Ehretia amoena* Klotzch. (rootbark); Capparidaceae: *Boscia salicifolia* Oliv. (leaves), *Maerua angolensis* DC. (bark), (leaves); Caricaceae: *Carica papaya* L. (green fruits), (bark); celastraceae: *Elasodendron sehlechteranum* Loes. (roots), (leaves); Combretaceae *Combretum zeyheri* Sond. (fruits), *Terminalia catappa* L. (leaves); Compositae:: *Vernonia hildebrandtii* Vatke. (leaves and stem), *V. cinerea* Less. (plant); Connaraceae: *Byrsocarpus orientalis* Bak. (plant); Dilleniceae: *Tetracera boiviniana* Baill. (rootbark); Ebenaceae: *Diospyros mespiliformis* Hochst. ex DC. (leaves); Euphorbiaceae: *Acalypha fruticosa* Forsk. (leaves), (roots), *Antidesma venosum* E. Mey. (rootbark), *Bridelia cathartica* Bertol. f. (leaves), *Euphorbia hirta* L. (Plant), *Fluggea virosa* Baill. (bark), *Phyllanthus reticulatus* Poir. (leaves), *Securinegaa virosa* Baill. (roots); Icacinaceae: *Pyrenacantha caurabassana* Baill. (tuber), (green fruits); Labiatae: *Hoslundia opposita* Vahl. (leaves), *Leonotis nepetaefolia* R. Br. (plant), Lauraceae: *Cassytha filiformis* L. (plant); Leguminosae; *Acacia mellifera* Vahl. (bark), *A. robusta* Burch. (rootbark), *A. senegal* Willd. (roots), *Adenanthera pavonina* L. (seeds), (leaves), *Bauhinia reticulata* DC. (plant), *Caesalpinia pulcherrima* Swartz. (flowers), (rootbark), *Cassia fistula* L. (bark), *C. obtusifolia* L. (plant), *C. occidentalis* L. (plant), *Desmodium* sp (plant). *Dichrostachys cenerea* Wight. Arn. (stem). *Peltophorum petocarpum* K. (roots), (bark), *Pongamia pinnata* P. (leaves and rootbark), *Pterocarpus angolensis* DC. (bark), *Stylosanthes fruticosa* Alston. (plant), Liliaceae: *Asparagus* sp. (plant); Loganiaceae: *Strychnos madagascarensis* Poir. (rootbark); Malvaceae: *Malvastrum coromandelianum* Garcke. (plant); *Sida cordifolia* L. (roots), *S. serratifolia* L. (plant), *S. spinosa* L. (roots), (leaves), Ochnaceae: *Brackenridgea Zanguebarica* Oliv. (rootbark); Rhamnaceae: *Zizyphus pubescens* Oliv. (stem); Rubiaceae: *Lamprothamnus zanguebaricus* Hiern. (leaves). Sapindaceae: *Allophylus rubifolius* Engl. (stem), *Deinbollia borbonica* R. (leaves); Solanaceae: *Withania somnifera* Dun. (plants); Sterculiaceae: *Dombeya shupangae* K. Schum. (bark), (leaves), *Melhania velutina* Forsk. (leaves), *Waltheria indica* L. (leaves); Tiliaceae: *Corchorus olitorius* L. (fruits and seeds) *Grewia stuhlmannii* K. Schum. (roots); *Triumfetta rhomboidea* Jacq. (bark and roots); Verbenaceae: *Lantana camara* L. (leaves) Verberiaceae: *Vitex* sp. (plant), (roots), Vitaceae: *Cissus rotundifolia* Vahl. (leaves).

incanum (fruit) and *Zizyphus pubescens* (leaves). Both of them had ++ antifungal activity. The root bark of *Harrisonia abyssinica*, made into a topical application, is a popular remedy for skin diseases and haemorrhoids in East Africa and elsewhere on the African continent. It is interesting to note that the extract of this plant had excellent antigonococcal activity (++++) [8] and also was highly active (++++) against *Staphylococcus aureus* in *in vitro* bioassay tests [7].

In general, *in vitro* antimycotic activity among the group A plants was relatively low i.e. +++ in 14 %, ++ in 57 % and + in 29 % of the plants found positive for antifungal activity. While the extracts showing high level of antimycotic activity came from plants in group B.

The majority of the extracts having activity against *T. mentagrophytes*, also inhibited the growth of *Candida albicans* but the activity was of a milder nature.

Further work on the isolation and identification of the constituents responsible for antifungal activity is in progress.

Acknowledgement. The authors wish to thank Mr. R. Wingfield for the identification of plants.

REFERENCES

1. E.P. Taylor, and P.F. D' Arcy, *Progress in Medicinal Chemistry* (Prenum, New York, 1961), p. 220.
2. A.O. Lucas and H.M. Gilles, *A Short Textbook of Preventive Medicine for the Tropics* (English University Press, London 1973), p. 127.
3. J.O. Kokawaro, *Medicinal Plants of East Africa* (East African Literature Bureau, Kampala, Nairobi, Dar es Salaam, 1976).
4. J. M. Watt, K.M. Breyer-Brandwijk, *Medicinal and Poisonous Plants of Southern and Eastern Africa* (Livingstone, London, 1962), second edition.
5. Information from local medicine men.
6. V. Beaman-Mbaya and S.I. Mohammed, *Antimicrobial Agents and Chemotherapy* (1976), pp. 920-24.
7. M.R. Khan, A.N. Sawhney, G. Ndaalio, M.H.H. Nkunya and H. Wevers, (in press) Part I.
8. A.N. Sawhney, M.R. Khan, G., Ndaalio, M.H.H. Nkunya and H. Wevers, (in press), Part II.