1111/. Chem. Pharll1. Med. J. Vol. 1(2), pp.145-147 (2004)

RESULTS OF RAPID ANTI MICROBIAL SENSITIVITY AND TOXICITY TEST OF THE EXTRACTS OF *SWERTIA CHlRATA, SYMPLOCOS RACEMOSA* AND *SOLANUM NIGRUM*

MansoorAhmad" Mehjabeen2, Noorjahan2, Noor Muhammad Shah" Manzoor Ahmad3, Hyder Raza2, Saeed Khan4 and Asif Bin Rehman2

Research Institute of Pharmaceutical Sciences, Depm1ment of Pharmacognosy, and Faculty of Phal7llacy, University of Karachi, Karachi-75270, Pakistan 2Department of Pharmacolog Faculty of Pharmacy, University of Karachi, Karachi-75270, Pakistan 3HEJ research Institute of Chemistry, University of Karachi, Karachi-75270, Pakistan 4Liaquat National Hospital, Karachi, Pakistan

ABSTRACT: An evaluation of anti microbial and cytotoxic effects of the plants extracts of *Solanum nigrum* (two varieties i.e. black and red fruits), *Swertia chirata, Symplocos racemosa* used as a medicinal agent in the cure of different ailment. The cytotoxic tests were carried ou~ on Artemia Salina (brine shrimp). The results showed that there was no positive lethality of any of the plant except the *Solanum nigrum* having black barrier/fruits (SNBS). LD50 i.e. 927.0293 *j.lg/ml* was found in upper toxic concentration, 258008.100 and lower toxic concentration is 231.0767.

The anti bacterial activity of crude extracts were studied *in vitro* on gram positive and gram negative bacteria *E. coli, Staphylococcus aureus, Pseudomonas aeruginosa, Klebsiella pneumoniae* while antifungal activity was foundona range of concentration between 20-700mg/disc of dry extract and fungal organisms, *Candida albicans* and *Aspergillus niger.* The crude extracts except SNBS were devoid of any toxicity on Artemia salina with in the range of antimicrobial concentration, suggesting that the action is selective on microorganism.

INTRODUCTION

Swertia chirata (Gentianaceae) is an erect annual herb (Anonymous 1995). The drug consist chiefly of the stem, which is a dark purplish brown color, flower is panicled small green. It is widely used to treat fever, malaria and liver diseases (Najumdar and Guha, 1993). In addition it is reported to have anti-inflammatory activity. Xanthone deriva-tives are known to possess significant anti-inflammatory activities (Najumdar and Guha, 1993). Several varieties of xanthone show potent anti-platelet, anticancer, antifungal, antimalarial and significant CNS stimulant effects. It is also used as bitter antistomachic, febrifuge, antihelmintic, diuretic, antiepileptic and for certain type of mental disorders (Najumdar and Guha, 1993).

Symplocos racemosa (Symplocaceae/ Styraceae) is a small tree with dark green leathery leaves. In indigenous system of medicines S. *race1170sa* is used for management of menstrual disorders and to provide firmness to spongy and bleeding gums. Its decoction is also used for the treatment of bowel complaints and ulcer. The astringent bark of S. *racemosa* is recommended for the treatment of eye diseases and uterine disorders (Jain. 1985).

The leaves and fruits of *Solanum nigrum* L. (Solanaceae) i.e. black night shade locally called Mako, are used medicinally (Anonymous). Reported constituents of S. *nig rum* are solanine, saponin, oil, dihydroxy stearic, palmitic and stearic acid. The barriers are oleogenous, bitter, pungent, laxative, alterative, aphrodisiac, tonic and diuretic. They are 6mm in diameter, globoseusuaUy found in purplish black, green (unripe), red and sometime yellow. Different varieties of S. *nigrum* found in other part of the world included South Africa, Germany and America. It is used by Europeans as a remedy for convulsions. In Southern Rhodesia the plant is used as African remedy for malaria, black water fever, dysenteries and other diseases. In Mouriti!Js a poultice of the plant is applied for the relief of abdominal pain and inflammation of the urinary bladder. It has been also used in the treatment of headaches, ulcers and wounds (Akhtar and Muhammad, 1989; Jain and Borthakur,. 1986). The fresh. young leaves contain Img/lOOg of ascorbic acid. The frpit has been used as a tonic in heart diseases, fever, diarrhea (Anonymous 1956). eye disease and in liver and other complaints. Freshly prepared fluidextract from all portions of plant has been recommended dropsy. heart disease, piles. gonoIThea. inflammatory swellings (Watt and Breyer-Brandwijk, 1962) and chronic cirrhosis of the liver and spleen (Nadkami, 1976).

Plants Name	*E. coli*	*K. pneumoniae*	*P. aeruginosa*	S. *aureus*
S. *chirata*	9mm	6mm	7mm	8mm
S. *racemosa*	7mm	6mm	6mm	8mm
SNBL	7mm	7mm	R	R
SNRL	R	9mm	8mm	8mm
SNRS	8mm	8mm	8mm	8mm
SNBS	7mm	9mm	8mm	6mm

Table 2 .
Antifungal Activity *of* Different Plant Crude Extracts

Name *of* plant	Zone *of* inhibition against *Candida albicans*	Zone *of* inhibition against *Aspergillus niger*
S. *chirata*	R	9mm
S. *racemosa*	R	9mm
S. *nig rum* (SNBS)	8mm	R
S. nigru111 (SNRS)	R	10mm
S. *nigrum* (SNBL)	8mm	9mm
S. *nigm111* (SNRL)	6mm	10mm

EXPERIMENT AL

Extraction of plant materials: S. *racemosa,* S. *chirata* and two varieties *of Solanum nigru111* having red and black fruits were used to investigate the anti microbial and cytotoxic activities. Two kilograms *of* each plant material Was crushed/cut into small pieces and soaked in ethanol (5 liters each) for 15 days. After this period material was filtered and the solvent was evaporated under reduced pressure on a rotary evaporator. A semisolid (gummy) extract was obtained. To this semisolid extract anti microbial and toxicity test were performed.

Antimicrobial activity of the crude extract: The pure cultures *of* bacteria *Escherichia coli* (gram negative). *Staphylococcus aureus* (gram positive) *Klebsiella pneumoniae* (gram negative), *Pseudomonas aemginosa* (gram negative) and fungi *Candida albicans* and *Aspergillus niger* were obtained from Liaquat National Hospital Karachi. The bacterial cultures were maintained on a nutrient broth medium incubating at 37°C for 48 hours, while fungi used in antimicrobial activity were stored on Sabouraud Dextrose agar in the refrigerator at 4°C before use.

Minimum inhibitory concentration (MIC) *of* extracts and pure compounds was performed by using standard protocols (Naqvi *et al.,* 1992).

Cytotoxicity assay: Solutions *of* the test sample were prepared by initially dissolving the 10 mg in 1 ml *of* DMSO and from this solution transfer 500 ,..d, 50 f.ll, and 5 f.ll to vials (3 vials/conc.) corresponding to 1000,100 and 10 f.lg/ml respectively. Dried it over night to cqpduct the brine shrimp lethality bioassay, maintained the brines shrimps eggs at 3TC in to the tank having continuous oxygen supply. A~low the eggs for maturation then place 30 larvae in to each vial containing 1000, 100 and 10 f.lg/ml *of* test samples. Observe the number *of* survived larvae in each vial (Meyer et aI, 1982; Maclaughlin *et aI.,* 1990; Amason *et al.,* 1989).

RESUL TS AND DISCUSSION

The antibacterial activity *of* crude extract *of* S. *chirata,* S. *racemosa,* S. *nigrum* (SNBL SNRL SNRS. SNBS) were performed on gram +ve and gram -ve microorganism. The results *of* this activity were measured in term *of* zone *of* inhibition. S. *chirata* showed maximum inhibitory response against *E. coli.* moderate activity against other selected microorganism where as S. *racemosa* showed highest response against S. *aureus.* No inhibitory zone was recorded against *P. aeruginosa* by S. *chirata,* S.

racemosa and SNBL but SNRL, SNRS and SNBS have positive results. In the data maximum response of the crude extract of SNRL was observed against *K. pneumoniae*.

The antifungal activity of all six crude extract was observed against *Candida albicans* and *Aspergillus niger*. SNBS SNBL and SNRL showed positive results against *Candida albicans* where as all the crude extracts except SNBS have significant activity against *Aspergillus niger*.

These antimicrobi.al results are a positive sign for further evaluation of the chemical constituents and can help in the future investigation of new antimicrobial ~~~. .

The toxicities of crude plant extracts to wards brine shrimps *(Artemia salina)* were performed. Brine shrimp larvae are commonly used for cytotoxicity assays (Pelka *et aI.,* 2000). These larvae are sensitive to toxic substances (Sultana *et al.,* 1999). The ratio between dead larvae (high motility in comparison to a. control with out any toxic substances is used to estimate the toxicity of the test solution. The results show that there is no positive lethality'for any of the plant extract except the SNBS which shows the positive cytotoxic effect with LDso 927.0293 Ilg/ml (upper toxic conc. is 258008.100 and lower toxic concentration is 231.0767). This assay on medicinal plants is carried out to evaluate the anticarcinogenic or antitumor activity. This is the relatively new technique and has some advantage for toxicity testing restorative materials because it can be quickly carried out at low cost.

References

1. Anonymous. Medicinal plants of Nepal for Ayurvedic drugs. (1995).
2. N. Najumdar, P. C. Guha. Indian medicinal plants *S1vertia chirata. J. Indian /11.5t. Sci,* 1993; 16A. 3409. 3. S. K. Jain. Medicinal plants. WBT, 1985; 140. 4. K. M. Nadkarni. The Indian Materia Medica. 3'd Edn.. Vol. I. Popular Prakashan, Bombay, 1976.
5. Anonymous. Medicinal plants (Indigenous and Exotic) palt V D. M. A.
6. M. S. Akhtar, M. Muhammad. Evaluation of the gastric antiulcerogenic effects of solanum species of economic importance, *Ann, Missouri Bot. Gard.* 1989; 75(3): 8.
7. Anon. 1956. The wealth of India: A dictionary of Indian Raw materials and Industrial products. Raw materials vol. IV: F-G. Council of scientific and industrial reach. New Dehli.
8. S.K.Jain, S.K. .Borthakur. 1986. Solanaceae in Indian Tradition Folklore and Medicine pp. 577-583.
9. J. M. Watt, M. G. Breyer- Brandwijk. 1962. Solanum nigrum L.pp 996-1000. in the medicinal and poisonous plants of southern and eastern Africa. .
10. M. M. Mackaen, A. M. Ali, N. H. Lajis, K. Kawazuo, Z. Hassall, M. Amran, M. Habsah, L. Y. Mooi, S.. M. Mohamed. *Journal of Ethno Pharmacology,* 2000: 72(3) pp.395-402.
11. M. Pelka, C. Danzl, W. Distler, A. Petschett. A new screening test for tqxicity testing of dental materials *Journal of Dentistry,* 2000: 28(5) pp. 0 341-345.
12. A. J. ~ultana, V. Ehtasharnulhaque, S. Qasim, U. ~ V. Ahmed. Cytotoxic activity of Marine macro algae on Artemia salina (brine shrimp). 1999, *Phytother. Res.* 13(4) pp.304-307.
13. S. B. S. Naqvi, D. Sheikh, K. Usmanghani, M. Shameel and M. R. Sheikh. *Pak. J. Pharm.Sci.* 1992; 5(2): 129-138.
14. B. N. Meyer, N. R. Ferrigni, J. E. Putnam, J. E. Jacobson, L.. B. P. E. Nicholas and 1. L. Mclaughlin. *Planta Medica.* 1982; 45: 31-34.
15. 1. L. Maclaughlin, C. J. Chang and D. L. Smith. Frontiers in Natural products (Ed. Atta-ur Rehman). 1990; 548-574.
16. J. T. Amason, B. 1. R. Philo gene and P. Moranel. *Insecticides of Plant origin.* 1989; 28-29.

Manuscript submitted on August 5,2004
Accepted for publication on August 26, 2004