



PHYTOCHEMICAL AND ANTIMICROBIAL STUDY ON *OROXYLUM INDICUM*

M.S. Rana^{1*}, N. S. Chowdhury¹, M. R. Karim¹ and G.M.M. Murshid²

¹Department of Pharmacy, Jahangirnagar University, Savar, Dhaka 1342, Bangladesh

²Pharmacy Discipline, Khulna University, Khulna 9208, Bangladesh

KUS -05/26-220905

Manuscript received: September 22, 2005; Accepted: October 16, 2006

Abstract: The root and stem barks of *Oroxylum indicum* was investigated for phytochemical constituents and antimicrobial activity. The powdered root and stem bark were successively extracted with ether, ethylacetate and ethyl alcohol. The extracts were examined to detect for various phytochemical constituents. Results showed the presence of carbohydrates, glycosides, alkaloids, flavonoids, tannins and steroids. Antimicrobial activities of the extracts were investigated. The ether and alcohol extracts were found to be active against almost all of the bacteria and fungus tested. The ethyl acetate extract of the root bark showed activity against four microorganisms- *C. albicans*, *S. dysenteriae*, *B. cereus* and *S. pyogenes* but that of stem bark showed no activity against any microorganism. All the activities were compared with the standard antibiotic by measuring the zone of inhibition.

Key words: *Oroxylum indicum*, phytochemical study, antimicrobial activity, agar diffusion method

Introduction

Oroxylum indicum (Linn) belonging to the family Bignoniaceae is an indigenous plant of Bangladesh and is widely distributed throughout the country. Literature review reveals that nine new naphthalene compounds together with four known compounds (Kiju *et al.*, 1994) and very small quantity of ellagic acid which were isolated from the root bark of the plant (Vasanth *et al.*, 1991). It was further reported that the plant extracts are widely used in cold fever, diarrhea, vomiting, gout, fungal infections (Joon *et al.*, 1998). In present work, an attempt was made to detect the presence of reported compounds by using various standard qualitative chemical tests and to look for possible presence of other chemical constituents in the root and stem bark of the extracts. Further, a simple and commonly used agar diffusion method was employed for antimicrobial investigation.

Materials and Methods

Sample collection and identification: The plant was collected from Khulna, Kustia, Natore, and Savar and identified by using standard taxonomical methods at National Herbarium, Dhaka. The identified plant parts were cut into small pieces separately and then dried in the sun and finally dried in a hot air oven at 50-60 °C

* Corresponding Author: Tel: +880-2-7708478~85; Fax: +880-2-7708069; e-mail: <sohel@juniv.edu>.

DOI: <https://doi.org/10.53808/KUS.2006.7.2.0526-L>

for 48 hours. After complete drying, the sample was crashed to coarse power separately with the help of a mechanical grinder and the powder was stored in a suitable container for extraction.

Preparation of the extract: The powdered plant material was extracted with ether, ethyl acetate and ethyl alcohol by the Soxhlet extractor fitted with a condenser. The crude extracts were made free from pigments (decolorized) and other impurities by filtering through activated charcoal. The decolorized extracts were then completely dried by vacuum rotary evaporator.

Phytochemical tests: Small amounts of dried, decolorized extracts were appropriately treated to prepare sample solution and then subjected to the specific phytochemical tests.

Determination of antimicrobial activities: One gram each of the dried extracts was dissolved in 5 ml of distilled water to prepare stock solution. As a solubilizing agent, few drops of Tween 80 (1%) were used to make the concentration of the solutions 200 mg ml⁻¹. From these solutions, 200µl of each sample were taken and applied into each hole or cup on the agar plate. To make a comparative study, amoxicillin and nystatin of same concentration was taken as a standard. Specific organisms were inoculated into 30 ml previously sterilized nutrient agar media, mixed thoroughly and transferred immediately to the sterile petridish in an aseptic condition. It was stored in an incubator for about 24 hours to allow the proper growth of microbes. Prepared sample solutions were applied to the corresponding cups or holes with the help of a micropipette. The plates were then allowed to stand to diffuse the sample solution into the antibiotic medium at room temperature for an hour. The plates were then incubated at 37 °C for overnight. After proper incubation, clear zones of inhibition around the point of application of sample solution were formed. These inhibition zones were measured by vernier scale and expressed in millimeter. For fungus, another type of culture medium was used and the same procedure was followed for the test (Bauer *et al.*, 1966).

Results

Qualitative phytochemical tests were performed for all the ether, ethyl acetate and ethyl alcohol extracts of the root and stem bark. The results of various chemical tests for the detection and/or identification of chemical constituents are summarized in the following table.

Antimicrobial activities of the root extracts of the plant were compared with the standard antibiotic amoxicillin and an antifungal drug nystatin by measuring the zone of inhibition in diameter. Results are presented in Table 2.

Discussion

As evident from the result of Table 1, the extracts of stem and root bark of *Oroxylum indicum* have been found to contain carbohydrates, alkaloids, steroids, proteins and tannins. Ether and alcohol extracts of the plant parts gave positive response to glucoside, flavonoid and glycoside tests but it found negative in case of ethylacetate extracts. However, all the extracts gave negative reaction to

Table 2. Comparative antimicrobial activities of the root bark extract of *Oroxylum indicum* and standard.

Code No.	Microorganisms	Diameter of inhibition zones (in mm)			
		EE	EaE	AE	Std.
F-01	<u>Fungus</u>				
	<i>Candida albicans</i>	22.0	15.5	22.2	22.2
	<u>Bacteria</u>				
B-01	<i>Salmonella typhi</i>	26.5	-	26.5	25.5
B-02	<i>Pseudomonas aeruginosa</i>	-	-	-	21.5
B-03	<i>Staphylococcus aureus</i>	29.0	-	30.1	26.0
B-04	<i>Vibrio cholerae</i>	21.8	-	22.0	22.0
B-05	<i>Shigella dysenteriae</i>	27.1	18	35.7	25.0
B-06	<i>Escherichia coli</i>	16.2	-	18.1	25.0
B-07	<i>Bacillus cereus</i>	27.5	16.1	32.2	29.0
B-08	<i>Streptococcus pyogenes</i>	36.0	25.2	37.5	38.2

EE = Ether Extract, EaE = Ethyl acetate Extract, AE = Alcohol Extract

Table 3. Antimicrobial activities of the stem bark extract of *Oroxylum indicum* and standard.

Code No.	Microorganisms	Diameter of inhibition zones (in mm)			
		EE	EaE	AE	Std.
F-01	<u>Fungus</u>				
	<i>Candida albicans</i>	20.0	-	22.2	22.2
	<u>Bacteria</u>				
B-01	<i>Salmonella typhi</i>	25.0	-	25.5	25.5
B-02	<i>Pseudomonas aeruginosa</i>	-	-	-	21.5
B-03	<i>Staphylococcus aureus</i>	29.0	-	29.8	26.0
B-04	<i>Vibrio cholerae</i>	21.2	-	21.5	22.0
B-05	<i>Shigella dysenteriae</i>	27.0	-	35.5	25.5
B-06	<i>Escherichia coli</i>	17.0	-	18.1	25.0
B-07	<i>Bacillus cereus</i>	25.5	-	30.0	29.0
B-08	<i>Streptococcus pyogenes</i>	36.0	-	37.5	38.2

EE = Ether Extract, EaE = Ethyl acetate Extract, AE = Alcohol Extract and Std. = Standard. Minus sign (-) indicates no antimicrobial activity. Std. = Standard. Minus sign (-) indicates no antimicrobial activity.

chemical constituents like saponin and anthraquinone glycoside tests. Further the qualitative test by TLC technique it was observed that the ether extracts traveled on silica gel plate giving several distinctive spots. In response to iodine vapor the spots visualized well and indicated the presence of different constituents. The R_f value calculated for the broad and uneven spot was approximately 0.40. The alcohol extracts gave three brownish colored spots in which one was broad and uneven round. For the broad and uneven spot, the R_f value was calculated as approximately 0.28. The ethylacetate extract of the plant parts exhibited a medium but uneven tailing almost half-way of the chromatogram. It gave also a small spot below the medium one. The observed spots indicated the presence of two different constituents. The medium spot may be for the presence of alkaloid. From these observations it can be indicated that the ether and alcohol extracts contain alkaloids, steroids and tannins which was in agreement with the previous reports (Kiju *et al.*, 1994; Grampurohit *et al.*, 1994).

Comparative antimicrobial activities of the root bark extract of *Oroxylum indicum* and standard was illustrated in Table 2. In this experiment, alcohol extract of *Oroxylum indicum* was the only extract which showed strongly significant sensitivity to the eight of the test organisms out of nine (Table 2). The highest zone of inhibition is 37.5 mm was recorded against *Streptococcus pyogenes*. Similar activities of the plant have been reported (Houghton *et al.*, 1997). If the antimicrobial activity of amoxicillin, which was taken as a standard, is compared with the antimicrobial activity of alcohol extract, the standard showed lower activity against *Shigella dysenteriae*, although it was found to be active against other test bacteria. Thus the alcohol extract of *Oroxylum indicum* showed broad spectrum antibacterial activity against a wide range of pathogenic bacteria including amoxicillin resistant *Shigella dysenteriae*. Significant antimicrobial activities towards the most test organisms were showed by ether extracts of the root bark. Ether extract failed to show its antibacterial activity against *Pseudomonas aeruginosa*. Ethyl acetate extract of the stem bark showed moderate antimicrobial activity against four organisms tested.

Table 3 demonstrated the antimicrobial activities of the stem bark of *Oroxylum indicum* and standard. The ether extract and alcohol extract of *Oroxylum indicum* inhibited the growth of fungi (*Candida albicans*). The maximum inhibition of growth (60%) was shown by the alcohol extract of root bark. In some cases, it was found that alcohol extract was very prominent to that of standard as the inhibition zone produced by nystatin was 22.2 mm while for alcohol extract it was also 22.2 mm. The antibacterial activities of alcohol extract and ether extract against *E. coli*, *S. dysenteriae*, *S. typhi* and *B. cereus* were highly promising. So, in case of diarrhoea and dysentery due to the aforementioned microbes can be cured by the application of ether extract and alcohol extract of *Oroxylum indicum* (Joon *et al.*, 1998). The alcohol extract and ether extract of both root and stem bark showed good activity against *Candida albicans* which is responsible for various infectious diseases. However, ethyl acetate extract of the stem bark showed no antimicrobial activity which can be explained by their phytochemical study. It was revealed that the bark extracts of the plant contain steroids, tannins, alkaloids (Table 1) which have potential role against different infectious diseases (Houghton *et al.*, 1997).

Conclusion

As evident from the above discussion, *Oroxylum indicum* contains important chemical substances that confer upon it as a medicinal agent which has broad spectrum antimicrobial activity. As apparent from our results and from other worker's reports, local uses of this plant in various

infectious diseases are not at much variance with their antimicrobial properties. This fact also indicates that the traditional uses of the plant are not scientifically baseless and therefore, the plant should be thoroughly investigated phytochemically to extensively exploit their medicinal and pharmaceutical potentialities.

References

- Bauer, A.W.; Kirby, W.M.M.; Sherris, J.C. and Turck, M. 1966. Antibiotic susceptibility testing by standardized single disc method. *American Journal of Clinical Pathophysiology*, 45: 493-496.
- Grampurohit, N.D.; Baichwal, M.R. and Jolly, C.I. 1994. Chemical constituents of the roots of *Oroxylum indicum* Vent. *Indian Journal of Natural Products*, 10(2): 8-12.
- Houghton, P.J.; Ali, R.M. and Azizol, M. 1997. Antimicrobial activity of extracts of some Bignoniaceae from Malaysia. *Pharmaceutical and Pharmacological Letters*, 7(2-3): 96-98.
- Joon, A.Y.; Joon, P.S.; Ha, C.D.; Chan, C.H. and Hiremath, I. 1998. Growth inhibitory responses of human intestinal bacteria to extracts from Indian and African plants. *Agricultural Chemistry and Biotechnology*, 41(1): 104-409.
- Kiju, H.; Habe, S.; Ishida, M. and Tomimori, T. 1994. Studies on the crude drugs and the naphthalene related compounds from the root bark of *Oroxylum indicum*. *Yakugaku Zasshi*, 114(7): 492-513.
- Vasanth, S.; Natarajan, M.; Sundaresan, R.; Bhima, R. and Kundu, A.B. 1991. Ellagic acid from *Oroxylum indicum* Vent. *Indian Drugs*, 28(2): 504-510.