BIOLOGICALLY ACTIVE PRINCIPLES ISOLATED FROM SALACIA OBLONGA WALL

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(Received on December 20, 1994)

Abstract: From the petroleum ether extract of the root bark of Salacia Oblonga wall, two biologically active fractions have been isolated by column and thin layer chromatography. The methanol eluted fraction of the extract absorbed on a column of silica gel at a concentration of 50 µg/ml showed 100 percent cytotoxicity on Ehrlich ascites tumour cells. The chloroform eluted fraction of the pet. ether extract and a fluorescent compound separated from it by TLC demonstrated about 60% and 76% hypoglycemic potency of an equal dose of tolbutamide (250 mg/kg) in albino rats. The results indicate the therapeutic importance of S. Oblonga wall.

Key	words:	Salacia oblonga
		fluorescent compound

INTRODUCTION

Salacia oblonga wall is found in South Indian forests and it is known as Ponkorandi in Kerala, because of its golden coloured roots. The root bark of this plant is used in gonorrohoea, rheumatism, and skin diseases (1). The aqueous and alcoholic extracts of the root bark of S. Oblonga have been shown to possess antidiabetic action (2, 3). The fractions of S. Oblonga extract were isolated and tested for cytotoxic and hypoglycemic activity.

METHODS

Root bark of S. Oblonga procured from the local market was sliced, sundried and powdered and a 500 g specimen was extracted exhaustively with petroleum ether (60-80°C) in a soxhlet. The extract was concentrated to 10 ml by distilling off the solvent and 2 ml of it was mixed with 10 g of silica gel (60-120 mesh size, Qualigens Fine Chemicals) and placed over a column of the same silica gel prepared as follows: Silica gel was soaked in petroleum ether (60-80°C) and packed in a glass column of 3 cm diameter to a height of 20 cm. The column was cytotoxicity

washed with petroleum ether. The extract was eluted successively with petroleum ether, petroleum ether-chloroform mixture (1:1), chloroform, acetone, methanol and water. The fractions were collected in 50 ml volumes. The collection of the elutes was as follows :

Eluting solvent	Fraction No. (50 ml each)	Nature of elutes	
Petroleum ether	1&2	Colourless	
Pet. ether- chloroform (1:1)	3&4	Yellow coloured	
Chloroform	5	Yellow coloured	
Acetone	6	Golden yellow coloured	
Acetone	odi taiy alt	Colourless	
Methanol	8	Pink coloured	
Water	9	Colourless	

The fraction 5 found to produce hypoglycemia was further purified by thin layer chromatography (TLC) using silical gel G coated on glass plates and a solvent system of toluene and acetone (95:5) (4). After running the samples

*Corresponding Author and present address : Professor, School of Medical Education, M.G. University, Medical College Campus, Kottayam - 686 008 (Kerala). for about half an hour the plates were dried in an oven and spots were watched under U.V. light and also visually. The visible and fluorescent bands were scrapped out and eluted with chloroform for the assay of the samples.

In vitro cytotoxicity on tumour cells : Cytotoxicity of the fractions was tested using Ehrlich ascites tumour cells (EAC). Different concentrations of each fraction were incubated with $1x10^6$ cells in phosphate buffered saline (pH-7.2) for 3h. The percentage of dead cells was determined by trypan blue exclusion method (5).

Hypoglycemic action : Albino rats (Sprague Dawley Strain) weighing 120-150 g were fed on standard diet and water *ad libitum*. Fasting blood samples were collected from tail-vein and rats were divided into groups of six each. One group was kept as control and similar groups were used as test groups for each isolated fraction and the standard antidiabetic agent tolbutamide.

Control rats were fed with 2 ml N. Saline. S. Oblonga fractions and tolbutamide (250 mg/ kg) suspended in 2 ml saline were administered orally to test groups using a gastric tube. Blood samples were collected at hrly intervals for four hrs and blood glucose was estimated (6). The results were statistically analysed by students' 't'-test.

RESULTS AND DISCUSSION

Only fraction 8 eluted by methanol gave a positive cytotoxic action on EAC. The death of cell was 75% at a concentration of 25 μ g/ml and 100% at 50 μ g/ml.

This suggests that the root bark of S. Oblonga contains an active principle that may inhibit the growth of tumours. However, further purification and identification of the

active principle and tests on other cancer cell lines and *in vivo* tests on solid tumour reduction are required to establish its anticancer action.

TABLE I: Hypoglycemic effects of the active fractions of the root bark of *Salacia Oblonga* Wall (250 mg/kg). Mean value of six rats ± S.D. are given for each group.

Groups and drugs	Blood glucose (mg/100 ml) Hours				ml)	Potency as percentage of tolbutamide
taphy The	0	1	2	3	4	3.52
Control Fraction 5 (eluted	100±7	95±4	94±5	96±5	98±4	-
by chloro- form)	99±5	96±6	86±4*	82±5**	80±6**	* 60
Fluores- cent com- pound Tolbuta-	94±5	82±6*	77±5**	75±6**	70±4.5*	* 76
mide	100±6	88±5*	82±6**	70±5**	60±4**	

*P<0.01; **P<0.001.

Fraction 5 eluted by chloroform alone had a significant hypoglycemic action in rats (Table I). This fraction on further separation on TLC gave one yellow band with an Rf of 0.62 and a blue fluorescent band visible under U.V. light with an Rf of 0.53. Of these the fluorescent compound produced a significant hypoglycemia. The efficacy on the basis of equivalent dose was less than tolbutamide. S. Oblonga appears to be a good source of biologically active principles which may be useful for producing hypoglycemia and for cytotoxicity to cancer cells.

ACKNOWLEDGEMENTS

The authors acknowledge with thanks Fr. Gabriel, Fr. Pius and Dr. R. Kuttan for their keen interest in this work.

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Indian J Physiol Pharmacol 1995; 39(4)

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