Pak. J. Agri. Sci.
24 (4) 235, 1987

PHYTOCHEMICAL SCREENING OF THE FRUIT OF *Caesalpinia crista* (Karanjwa), *Melia azedarach* (Bakain) AND ROOTS OF *Saussurea lappa* (Qust-e-Shireen)

M. S. Akhtar and Nosheena Farah

Department of Physiology and Pharmacology, University of Agriculture, Faisalabad.

ABSTRACT

The fruit of *Caesalpinia crista* (Karanjwa), *Melia azedarach* (Bakain) and roots of *Saussurea lappa* (Qust-e-Shireen) were found to exert anthelmintic effect in animals. In an effort to fractionate the active principle of these plant drugs, preliminary phytochemical screening has been done. The results showed that fruit of *Caesalpinia crista* contained 0.02% glycosides and 0.001% saponins but no alkaloids, anthraquinones, cardiac glycosides and flavonoids. The fruit of *M. azedarach* contained 0.1% glycosides and 0.01% bounded anthraquinones. The alkaloids, cardiac glycosides, flavonoids and saponins could not be detected in this fruit. However, *S. lappa* roots were observed to contain 0.04 % alkaloids, 0.01 % glycosides and 0.003 % cardiac glycosides, while anthraquinones, flavonoids and saponins were nil.

INTRODUCTION

The fruit of *Caesalpinia crista* (Leguminosae) *Melia azedarach* (Meliaceae) and the roots of *Saussurea lappa* (Compositae) locally known as "Karanjwa", "Bakain" and "Qust-e-Shireen", respectively, have been quite commonly used in "Unani" medicine (Nadkrani, 1954) for treatment of various diseases of man and animals.

The medicinal properties ascribed to these plants included anthelmintic, febrifuge, antiphlegmatic, antiflatulant, etc. In addition, medicinal plants have been used to cure nervous problems, skin diseases, cough, rheumatism, chronic fever, eczema and dyspaepsia (Chopra *et al.*, 1956; Ikram and Hussain 1978). Oral administration of plants in powdered form and their extracts in water and

methanol were found to exert anthelmintic actions in animals (Riffat and Akhtar, 1984). Therefore, preliminary phytochemical studies have been carried out in an effort to detect the compounds responsible for the reported pharmacological property in the plant under investigation.

MATERIALS AND METHODS

Plants and Chemicals Used

The seeds of *Caesalpinia crista* (Karanjwa), fruit of *Melia azedarach* (Bakain) and the roots of *Saussurea lappa* (Qust-e-Shireen) were purchased from local herbal dealers. All the chemicals used were of analytical grade prepared by E. Merck, Darmstadt, West Germany or B D.H. Laboratories, Poole, England

Experimental Procedure

Detection of alkaloids: The presence of alkaloids was checked by the Dragendroff's reagent (Harborne, 1963), Mayer's reagent (Mayer, 1963) and Wagner's reagent (Jenkins *et al.*, 1967). Then extracts were prepared for the detection of alkaloids by using the following methods:

Culvenor and Fitzyerald (1963): Five grams of plant material were slurred with 1 g of sand and 10 ml of chloroform. Then added 5 ml of 2N H_2SO_4 and filtered. The filtrate was made alkaline (pH = 8) and centrifuged for 10 minutes. The presence of turbidity with any one or all of the three detection reagents already mentioned was taken as an index for the presence of alkaloids.

Brain and Turner (1975): Ten grams of plant material were boiled with 100 ml of acidified ethanol and cooled. Then it was centrifuged, made alkaline (pH = 9) and extracted with 50 ml of chloroform. The chloroform layer was extracted with dilute hydrochloric acid, which in case of positive results gave orange precipitates with Dragendroff's reagent, greenish white in case of Mayer's reagent and reddish brown precipitates in case of Wagner's reagent.

Isolation and quantitative analysis of alkaloids: The method of Brain and Turner (1975) was adopted for this purpose. Ten grams of powdered plant material was shaken with 100 ml of 10% NH_4OH for 10 minutes. The filtrate was extracted with 50 ml of chloroform and to chloroform layer were added 50 ml of 50% Hcl. The acidic layer was neutralized with 10% NH_4OH (pH = 7) and again extracted with 50 ml of chloroform. The chloroform layer was

evaporated to dryness. The percentage of alkaloids was calculated on weight basis.

Detection and isolation of glycosides: The glycosides were extracted for the detection of glycosides by using the Brain and Turner (1975) method in which 10 grams of the plant material were boiled with 30 ml of 70% ethanol for 5 minutes, filtered and added concentrated lead acetate solution till no further precipitation occurred. The excess of lead acetate was removed by passing H_2S gas and filtered. The filtrate was concentrated to 5 ml and tested for the presence of reducing sugars by Fehling's (Harborne, 1963) and Benedict's solutions (Adam *et al.*, 1970).

Detection and isolation of cardiac glycosides: Using the method of Brain and Turner (1975), 10 g of powdered plant material was boiled with 100 ml of 70% ethanol for 5 minutes and filtered: diluted the filtrate with twice of its volume of distilled water and added 10 ml of saturated lead acetate. The excess of lead acetate was removed by 6.3% Na_2So_4 solution. Extracted the filtrate with 50 ml of chloroform. The chloroform was evaporated and dissolved the residue in 3 ml of 3.5% $FeCl_3$ prepared in acetic acid. Carefully added 1.5 ml of concentrated H_2SO_4 along the walls of test tube. A brown colour at interface and a pale green colour in the upper layer were taken as indicators of cardiac glycosides.

Detection of anthraquinones: Extracted 10 g plant drug with 50 ml of hot water, filtered, cooled and extracted with 20 ml of CCl_4. The CCl_4 layer was washed with 20 ml of water and shaked with 10 ml of 5% NH_4OH. A pink to cherry colour in the ammoniacal layer indicated the free anthraquinones. Another 10 g of plant were hydrolysed with 20 ml of 10% $FeCl_3$ for 10 minutes, cooled the filtrate and treated as described above. A more intense red colouration marked the presence of bounded anthraquinones (Brain and Turner, 1975).

Isolation of anthraquinones: Following the method of Brain and Turner (1975), 10 g of powdered plant was macerated with 50 ml of chloroform water (0.25%) for 8 hours and filtered. Heated the liquid at 80°C for 3 minutes to denature proteins and allowed to stand for 24 hours. Evaporated the filtrate under reduced pressure at a temperature not greater than 60°C to 10 ml and calculated the percentage of anthraquinones.

Detection of saponins: Extracted 10 g of plant material with 50 ml hot water, retained the filtrate and added 5 ml of 1.8% NaCl solution to each of two test tubes. To one of them added 5 ml of water and to the other 5 ml of the above filtrate, Added 8 drops of blood in both test tubes. Haemolysis in tube containing extract but not in control tube indicated the presence of saponins.

Isolation of saponins: Refluxed 5 g of plant material with 100 ml of 2N HCl for 2 hours and filtered. The residue was neutralized by passing NH_4OH (4%). Dried the residue at 60°C for 16 hours. Extracted the residue with petroleum ether (40° - 60°C) in Soxhlet apparatus for 24 hours. Reduced the solvent to 30 ml and placed inacool place (Brain and Turner, 1975).

RESULTS AND DISCUSSION

The data given in Table 1 showed that fruit of *C. crista* did not contain alkaloids, anthraquinones, cardiac glycosides and flavonoids. Instead the fruit had 0.02% glycosides and 0.001% saponins. The presence of glycosides (bonducin) and saponins was also earlier reported by Chopra *et al.* (1956). The anthelmintic activity of the *C. crista* may be due to the presence of glycosides and or saponins for which studies are in progress.

The screening of *M. azedarach* showed that it contained 0.1% glycosides and 0.01% bounded anthraquinones. Srivastava (1986) described the

Table 1. *Phytochemical constituents of caesalpinia crista (fruit), melia azedarach (fruit) and saussurea lappa (roots)*

Chemical constituents	*Caesalpinia crista*		*Melia azedarach*		*Saussurea lappa*	
	Present or not	Percent yield	Present or not	Percent yield	Present or not	Percent yield
1. Alkaloids	—	—	—	—	+	0.04
2. Glycosides	+	0.02	+	0.1	+	0.01
3. Cardiac glycosides	—	—	—	—	+	0.003
4. Flavonoids	—	—	—	—	—	—
5. Saponins	+	0.001	—	—	—	—
6. Anthraquinones						
Free :	—	—	+	0.01	—	—
Bounded :	—	—	—	—	—	—

chemical structure of limonoid, a glycoside isolated from the seeds of *M. azedarach* plant as 6-acetoxy-11-∝-hydroxy-7-oxo-14 B, 15 B-epoxymeliacin-1,5-diene-3-0-∝-L-rhamnopyranoside. The above mentioned glycoside was observed to possess antibacterial activity against several microorganisms. The presence of bounded anthraquinones in the fruit of *M. azedarach* is also in agreement with the findings of Srivastava and Mumta (1980). The new anthraquinone glycosides were isolated from its fruit and were characterized as 1, 8-dihydroxy-2- methylanthraquinone-3-0-B-D-galactopyranoside and 1, 5-dihydroxy-8-methoxy-2-methylanthraquinone-3-0-∝-L-rhamnopyranoside from chemical and spectral evidences by these workers.

The phyto-analysis of *S. lappa* roots revealed the presence of alkaloids, glycosides and cardiac glycosides in them. The *S. lappa* roots contained 0.40% alkaloids, 0.0 14% glycosides and 0.003% cardiac glycosides. The other components such as flavonoids, saponins and anthraquinones were, however, found to be absent. Earlier Nadkarni (1954) also reported the presence of alkaloids, glycosides and cardiac glycosides in the roots of *S. lappa*. Thus, the anthelmintic action of *S. lappa* roots (Qust-e-Shireen) may be due to any one or all of its three phytocomponents. It is hoped that further pharmacological/chemical studies would reveal their real active chemical principles or fractions responsible for the anthelmintic actions of these ethanomedicines commonly used in the eastern (Unani and Ayurvedic) medicine

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