



Short Communication

Cytotoxicity and antinociceptive activity of *Jasminum sambac* leaves

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SUMMARY

The ethanolic extract of *Jasminum sambac* leaves were tested for its cytotoxicity and possible antinociceptive activity in experimental animals. The extract showed potent cytotoxic activity in brine shrimp lethality assay and the LC₅₀ was found only 25 mg/ml. The extract significantly and dose dependently inhibited the acetic acid induced writhing in mice (56.83%, $P < 0.001$ and 43.17%, $P < 0.001$ for 500 and 250 mg/kg body weight, respectively). The results supported its traditional uses.

Key words: *Jasminum sambac*; Cytotoxicity; Antinociceptive; Brine shrimp

INTRODUCTION

Jasminum sambac (Family: Oleaceae), commonly known as 'Beliphul gachh', is a scandent shrub with opposite leaves pubescent branches, fragrant flowers, grown as an ornamental plant in garden all over the country (Ghani, 2003). Traditionally the aerial parts are used as CNS depressant, hypotensive and in treating insanity and weakness of sight. Root is used as an emmenagogue and leaves extract is used against indolent ulcer and breast tumours (Ghani, 2003). The plant has been reported to contain essential oil linalool, cis-jasmone, leaves contain iridoid glycosides and flower buds possess b-primeveroside and b-rutinoside glycosides (Junji *et al.*, 1995; Ghani, 2003).

As a part of our on-going pharmacological screening of randomly selected Bangladeshi medicinal plants (Shilpi *et al.*, 2004; Uddin *et al.*, 2004, 2005), we now report on the investigation of cytotoxicity and antinociceptive activity of *Jasminum sambac* leaves extract.

MATERIALS AND METHODS

Plant material

Leaves of *J. sambac* were collected from the Khanjahan Ali Hall, Khulna University, Khulna-9208, Bangladesh at the August 2005 when the plant was fully flowered and identified by experts of the Bangladesh National Herbarium, Dhaka, Bangladesh. Voucher specimens (DACB. 31262) representing the collections have been deposited in the Bangladesh National Herbarium, Dhaka, Bangladesh.

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Extraction

Shade-dried and ground leaves (300 g) were extracted by maceration over 24 - 72 h using 90% ethanol (EtOH) at room temperature. The ethanolic extract of *Jasminum sambac* (EEJS) was filtered and air dried. The yield was approximately 5% w/w on dry weight basis.

Animals

Swiss albino mice of either sex (20 - 25 g) were obtained from the Animal house, Pharmacy Discipline, Khulna University, Khulna. The animals were housed under standard laboratory conditions (relative humidity 55 - 65%, room temperature $23.0 \pm 2.0^\circ\text{C}$ and 12 h light-dark cycle). The animals were fed standard diet and water *ad libitum*.

Brine shrimp lethality assay for general toxicity

The method of Meyer *et al.* (1982) was adopted to study the general toxicity of the EEJS. The brine shrimp eggs were hatched in a conical flask containing brine shrimp medium (300 ml). The flasks were well aerated with the aid of an air pump, and kept in a water bath at 29 - 30°C. A bright light was left on. The nauplii hatched within 48 h. The extracts were dissolved in DMSO to obtain a concentration of 10, 20, 40, 80 and 160 µg/ml in 5% DMSO. Each extract preparation was dispensed into clean test tubes in 10 ml volumes and tested in duplicates. The concentration of DMSO in the vials was kept below 10 µl/ml. For control, same procedure was followed except test samples. After marking the test tubes properly, 10 living shrimps were added to each of the 20 vials with the help of a Pasteur pipette. The test tube containing the sample and control were then incubated at 29°C for 24 h in a water bath, after which each tube was examined and the surviving brine shrimps counted and recorded. From this, the percentage of mortality was calculated at each concentration.

Antinociceptive activity study using acetic acid induced writhing assay

The method of Uddin *et al.* (2005) was adopted with minor modification. The animals were orally fed with the extracts, vehicles (for control groups) at the specified doses (500 mg/kg body weight). Forty five minutes after administration of the extract and the vehicle, each animal was given 0.7% (v/v) solution of acetic acid (0.1 ml/10 g body weight) interperitoneally (i.p.) to induce abdominal contractions or writhing. Five minutes after the administration of acetic acid, the number of writhing for each animal was counted for 15 min. The number of writhings in the control was taken as 100% and percent inhibition was calculated as follows:

$$\% \text{ Inhibition of writhing} = 100 - (\text{treated mean} / \text{control mean}) \times 100$$

For comparison, the same experiment was carried out with a positive control group treated orally with Diclofenac-Na (Square Pharmaceuticals Ltd., Bangladesh) at the dose of 25 mg/kg body weights.

Statistical analysis

All data were expressed as mean \pm S.E.M. The Student's *t*-test was used to analyze data obtained from *in vivo* experiments.

RESULTS AND DISCUSSION

The general toxicity of the leaves extract EEJS was investigated by brine shrimp lethality assay method. The brine shrimp lethality assay is low cost assay method and indicator of toxicity. The leaves extract EEJS showed potent toxicity in the brine shrimp lethality assay and the IC_{50} value was 25 µg/ml (Table 1).

In acetic acid induced writhing test, the extract EEJS significantly and dose dependently suppressed the frequency of acetic acid induced writhing in mice. At the dose 250 mg/kg body weight the extract EEJS showed 43.17% writhing inhibition ($P < 0.001$) where as at 500 mg/kg body weight

Table 1. Brine shrimp lethality bioassay of *J. sambac* leaves extract (EEJS)

Test sample	Conc. of extract. ($\mu\text{g/ml}$)	Log (Conc.)	No. of alive shrimp	% mortality	LC ₅₀ ($\mu\text{g/ml}$)
EEJS	10	1	08	20	25
	20	1.3	06	40	
	40	1.6	01	90	
	80	1.9	0	100	
	160	2.2	0	100	

Table 2. Effect of the ethanolic extract of *J. sambac* leaves (EEJS) on acetic acid induced writhing in mice

Treatment	Dose ^a (mg/kg, p.o.)	Writhings ^b	Writhing Inhibition (%)
Control (1% Tween 80, 10 ml/kg, p.o.)	-	27.8 \pm 1.85	-
Diclofenac sodium	25	6.2 \pm 0.74*	77.7
EEJS	250	15.8 \pm 1.52*	43.17
	500	7.2 \pm 1.39*	56.83

^aAdministered 45 min before 0.7% acetic acid administration (ml/kg, i.p.). ^bCounted for 15 min, starting 5 min after acetic acid administration; values are mean \pm S.E. * $P < 0.001$ vs. control, Student's *t*-test ($n = 5$).

produced 56.83% writhing inhibition ($P < 0.001$), where the standard drug diclofenac-Na showed 75.2% writhing inhibition and all the results are statistically significant (Table 2).

From the study we found that the EEJS showed potent cytotoxicity and possesses antinociceptive activity with a dose dependant manner. Further study could be carried out to evaluate the antinociceptive activity and cytotoxicity of the extract.

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