Iranian Journal of Pharmaceutical Sciences
Summer 2009: 5(3): 151-156
ijps.sums.ac.ir

Original Article

Evaluation of Anti-inflammatory Activity of Methanolic Extract of *Solanum nigrum* (Solanaceae)

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Abstract

The methanolic extract of whole plants of *Solanum nigrum* L. was investigated for anti-inflammatory activity on the experimental animal models. The methanolic extract at a concentration of 100 $mg.kg^{-1}$ and 200 $mg.kg^{-1}$, *p.o.* showed the significant dose dependent anti-inflammatory activity in carrageenin and egg white induced hind paw oedema in rats. Anti-inflammatory activity of the tested extract was comparable with that of the standard drug indomethacin (10 $mg.kg^{-1}$) and cyproheptadine (8 $mg.kg^{-1}$). The results lend support to the traditional use of *Solanum nigrum* in the treatment of inflammatory diseases.

Keywords: Anti-inflammatory activity; *Solanum nigrum*; Solanaceae.
Received: November 4, 2008; *Accepted:* April 6, 2009

1. Introduction

The plant *Solanum nigrum* is well known in Tamil Munatakali and Hindi Makoi. A herbaceous or suffrutescent weed, 30-45 cm high, found throughout India in dry parts, up to an elevation of 2,100 m. Leaves ovate or oblong, sinuate-toothed or lobed, narrowed at both ends; flowers white, in drooping umbel-like 3-8 flowered clusters; berries red, yellow or black, round; seeds dicoid, smooth, yellow, minutely pitted [1]. The use of *S. nigrum* as stock for tomatoes to counteract the heat in North India has been suggested. The herb has antiseptic and antidysenteric properties and is given internally for cardalgia and gripe. An infusion of the plant is used as an enema in infants having abdominal upsets. It is a household remedy for anthrax pustules and is applied locally. The plant is also credited with emollient, diuretic and laxative properties and its decoction is regarded as an antispasmodic and narcotic. Freshly prepared extract of the plant is effective in the treatment

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of cirrhosis of the liver, and also serves as an antidote to opium poisoning. An alcoholic extract of leaves is active against *Staphylococcus aureus* and *Escherichia coli*. Infusions or decoctions of the plant after transient stimulation, depress the central nervous system and the reflexes of the spinal cord. Leaves are used in the treatment of scrofulous dyscrasias, and are said to produce diaphoresis when in overdose; they are said to produce diaphoresis when in over dose; they also cause nausea, purging and nervous disturbances. In China, leaves are applied to wounds and sores. Berries are considered to possess tonic, diuretic and cathartic properties and are useful in anasarca and heart diseases. They are a domestic remedy for fevers, diarrhoea, ulcers and eye troubles. Aqueous extract of the ripe fruit inhibit choline esterase activity of human plasma [1]. The aim of the present work is to evaluate the anti inflammatory activity of *S. nigrum* in experimental animal models.

2. Materials and methods

2.1. Plant material

The whole plants of *S. nigrum* was collected from the Tirunelveli District, Tamilnadu, India during October 2005. The sample was identified and the voucher specimen was deposited at S. A. Raja Pharmacy College, Raja Nagar, Vadakkangulam - 627116, Tamilnadu, India. The whole plants of *S. nigrum* were dried under shade, pulverized by a mechanical grinder and passed through sieve to get the fine powder.

Table 1. Preliminary phytochemical groups test for the methanolic extracts of whole plants of *S. nigrum*.

Phytochemical tests	Result of the test
Alkaloids	+
Steroids	+
Triterpenoids	+
Amino acids	+
Flavonoids	+
Gums	+
Reducing sugars	+
Tannins	+
Saponins	+

2.2. Preparation of extracts

The powdered whole plants of *S. nigrum* were successively macerated in 95% methanol as solvent for 72 h with occasional shaking at room temperature. The extract was collected in a conical flask, filtered through Whatman no.1 fitter paper and the filtrate was evaporated to dryness under reduced pressure. The yield of the prepared extract was around 4.26% w/w.

2.3. Animals

Albino Wister rats of either sex (160-180 g) were used for the study of anti-inflammatory activities. They are housed for at least one week before starting experiment in standard plastic cages at room temperature. The animals had free access to standard food in pellets and tap water.

2.4. Preliminary phytochemical group test

The preliminary phytochemical group test of the methanolic extracts of whole plants of *S. nigrum* was performed by the standard methods [2-5].

2.5. Anti-inflammatory activity

2.5.1. Carrageenin-induced rat paw oedema

The rats weighing 160-180 g were divided into four groups, each group consisting of six animals. Paw oedema was induced by subplantar injection of 0.1 ml of freshly prepared 1% carrageenin suspension into the right hind paw of each rat. The paw volumes were measured using a plethysmometer before as well as 60, 120,180 and 240 min. after the injection of carrageenin [6]. The methanol extracts of whole plants of *S. nigrum* at 100 and 200 $mg.kg^{-1}$ were administered orally to first two groups of rats. The third and fourth group of rats received 5 $ml.kg^{-1}$ propylene glycol as vehicle control or 10 $mg.kg^{-1}$

Table 2. Inhibitory effect of methanolic extract of whole plants of *S. nigrum* (MESN) against carrageenin induced paw oedema in albino rats.

Treatment	% Increase in paw volume, mean±S.E (n=6) Post insult time of assay in min.					% Inhibition in paw volume
	0	60	120	180	240	
Propylene glycol (5 ml.kg^{-1})	39.91±1.53	69.32±3.12	97.83±8.13	108.59±9.09	109.81±8.33	-
MESN (100 mg.kg^{-1})	28.72±1.86	49.32±4.50	72.40±4.50	72.40±6.90*	66.87±6.12*	38.41
MESN (200 mg.kg^{-1})	30.25±2.07	47.62±4.20	61.54±5.40	59.93±4.70*	56.26±5.10*	44.81
Indomethacin (10 mg.kg^{-1})	27.90±0.92	33.80±1.83	38.80±2.32	55.90±3.21*	58.82±3.92*	48.52

*p<0.001 *vs* control by students 't' test.

indomethacin as drug control, respectively, for comparative pharmacological assessment. Test drugs and vehicle were given 1 h before the injection of carrageenin. The relative potency of the drugs under investigations was calculated based upon the percentage inhibition of the inflammation.

2.5.2. Egg white induced hind paw oedema

Albino Wistar rats of either sex weighing about 160-180 g were divided into four groups of six animals each. The methanol extracts of whole plants of *S. nigrum* at 100 and 200 mg.kg^{-1} were administered orally to first two groups of rats. The third and fourth group of rats received 5 ml.kg^{-1} propylene glycol as vehicle control or 8 mg.kg^{-1} cyproheptadine as drug control, respectively, for comparative pharmacological assessment. All the drugs and vehicle were given one hour prior to the study. Freshly taken egg white (0.1 ml) was injected into the sub plantar tissue of the left hind paw of the rat. The volumes of the injected paws were measured at 0, 60, 120, 180 and 240 min. using a plethysmometer. The percentage of increase in paw oedema of the treated group was compared with that of the control and the inhibitory effects of the drugs were studied [7]. Percentage inhibitions were calculated for both models by using the following formula:

V_C-V_T/V_C×100

V_C=Control (% increase in paw volume in 3rd h)

V_T=Test (% increase in paw volume in 3rd h)

2.6. Statistical analysis

The results were expressed as mean±S.E and the significance were evaluated by student's t-test compared with control [8].

3. Results

3.1. Preliminary phytochemical group tests

Preliminary phytochemical group test were performed by using standard protocol and the results are presented in Table 1. The result showed that the various phytoconstituents present in methanolic extracts of whole plants of *S. nigrum* like steroids, triterpenoids, alkaloids flavanoids, reducing sugar, tannins, gums and saponins.

3.2. Anti-inflammatory activity

The anti-inflammatory potential of the methanol extracts of whole plants of *S. nigrum* was investigated using carrageenin-induced rat paw oedema and egg white induced hind paw oedema methods. The results of methanolic extracts of whole plants of *S. nigrum* in carrageen induced hind paw oedema are presented in Table 2. The results revealed that the methanolic extracts of whole plants of *S. nigrum* at 100 and 200 mg.kg^{-1} exhibited maximum inhibition was 38.412% and 44.81%, respectively in carrageen induced hind paw oedema; while idomethacin at showed 48.52% inhibition of oedema after 3 h of drug treatment (Table 2). The results of egg white induced hind paw oedema test showed that the oedema suppression by whole plant extract of *S. nigrum* at 100 and 200

Table 3. Anti-inflammatory activity of methanolic extract of whole plants of *S. nigrum* (MESN) against egg white induced paw oedema in albino rats.

Treatment	% Increase in paw volume Mean±S.E (n=6) Post insult time of assay in minutes					% Inhibition in paw volume
	0	60	120	180	240	
Propylene glycol (5 ml.kg^{-1})	45.90±3.70	81.20±5.2	94.3±5.70	92.50±2.30	90.10±3.40	-
MEEP (100 mg.kg^{-1})	35.72±2.69	52.63±3.9	78.3±5.29	69.24±5.82*	61.36±5.50*	25.15
MEEP (200 mg.kg^{-1})	33.19±1.78	49.58±3.6	69.42±4.2	62.31±3.83**	59.67±4.40**	32.63
Cyproheptadine (8 mg.kg^{-1})	28.60±1.80	44.8±4.2	49.6±3.10	47.80±2.30**	42.70±2.80**	48.32

*p<0.001 *vs* control by students 't' test; ** I<0.001 *vs* control by students 't' test.

mg.kg^{-1} was 25.15% and 32.63%, respectively; whereas cyproheptadine (8 mg.kg^{-1}) produced 48.32% inhibition (Table 3). Anti-inflammatory intensity produced by methanol extracts of whole plants of *S. nigrum* is comparable to that of the standard drugs indomethacin and cyproheptadine used in this study.

4. Discussion and conclusion

The phytochemical analysis of the plant extract using methods described indicated the presence of steroids, triterpenoids, alkaloids, flavanoids, tannins, reducing sugar gums and saponins. The earlier studies had indicated the use of egg-albumin as a phlogistic agent in causing oedema in rat hind paw. Carrageenin- induced rat paw oedema and egg white induced hind paw oedema methods are suitable for screen agents for anti-inflammatory activity which are frequently used to assess the anti-oedematous effect of natural products [9, 10].

Several inflammatory mediators like complement, histamine, kinins, prostaglandins and pro-inflammatory cytokines have been suggested to play a role in the mechanism of inflammation [11, 12]. It is assumed that at least some of these mediators are subjects of inhibition by the methanol extracts of whole plants of *S. nigrum*.

Oedema which develops after carrageenin inflammation is a biphasic event [13]. The initial phase is attributed to the release of histamine and serotonin. The oedema maintained between the first and the second phase is due to kinin like substances [14]. It has been reported that the egg white acts prominently on the mast cells. Oedema induced by it appears to be mediated by histamine and serotonin. Inflammatory processes in which mast cells are prominently involved are inhibited by antihistaminic and anti-serotonin compounds in the rat. The anti-oedematous effect showed by methanol extracts of whole plants of *S. nigrum* was significant during the first phase of oedema development and significantly maintained in the second phase of the oedema development, suggesting an inhibitory effect on the release of active pain substance such as histamine, serotonin, polypeptides or prostaglandins.

The steroids, alkaloids and triterpenoids present in the extract may be responsible for this anti-oedematous effect. Thus, further work is essential to fractionate, purify and identify the active principle(s) presenting this extract, as well as to understand the precise mechanism of action in anti-inflammatory activities by the methanol extracts of whole plants of *S. nigrum* .

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