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Research Article

**EXTRACTION, PHYTOCHEMICAL SCREENING AND
EVALUATION OF ANALGESIC & ANTIPYRETIC ACTIVITY
OF HYDROALCOHOLIC LEAF EXTRACT OF SOLENUM
VIRGINIANUM**¹Kartik Verma, ²Dr. Deepak Basedia, ³Dr. B. K. Dubey¹TIT College of Pharmacy, Bhopal (M.P.)**Article Received:** February 2022**Accepted:** February 2022**Published:** March 2022**Abstract:**

Solanum virginianum used by the local people as folk medicines in treating throat infections and other inflammatory problems. The fruits are known for several medicinal uses like anthelmintic, antipyretic, laxative, anti-inflammatory, anti-asthmatic and aphrodisiac activities. The fruit paste is applied externally to the affected area for treating pimples and swellings. The various parts of the plant are reputed in indigenous Hindu Medicine to have high medicinal value in various diseases like cough, asthma, fever, heart disease etc. This study was designed to evaluate different in vitro biological activities like antioxidant activity, antimicrobial activity and in vivo anti-pyretic and analgesic activities of hydroalcoholic extract of leaves of *Solanum virginianum*. The effect of hydroalcoholic extract of leaves of *Solanum virginianum* were determined after administration at two dose levels (100 and 200 mg/kg b.w.) in yeast induced pyrexia and tail immersion in rats. Our findings suggest a medicinal plant with multiple therapeutic properties that could serve as an antipyretic drug. To elucidate the exact mechanism of action of these active constituents, further investigation is required.

Key words: *Solanum virginianum*, Hydroalcoholic extract, Analgesic, Antipyretic Activity**Corresponding author:****Kartik Verma**

TIT College of Pharmacy, Bhopal (M.P.)

karthikverma.220@hotmail.com

QR code



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INTRODUCTION:

Herbal medicines and their preparations have been widely used traditionally, for the thousands of years in developing and developed countries owing to its natural origin and lesser side effects or dissatisfaction with the results of synthetic drugs. One of the characteristics of oriental herbal medicine preparations is that all the herbal medicines, either presenting as single herbs or as collections of herbs in composite formulae (Balammal *et al.*, 2012). The traditional preparations comprise medicinal plants, minerals, organic matter, etc. Herbal drugs constitute mainly those traditional medicines which primarily use medicinal plant preparations for therapy (Pal and Shukla, 2003). These drugs are made from renewable resources of raw materials by eco-friendly processes and will bring economic prosperity to the masses growing these raw materials (Kashaw *et al.*, 2011).

India is known as the “Emporium of Medicinal plants” due to availability of several thousands of medicinal plants in the different bioclimatic zones (Prabhu *et al.*, 2012). Medicinal plants continue to provide valuable therapeutic agents, both in modern medicine and in traditional systems of medicine. Attention is being focused on the investigation of efficacy of plant based drugs used in the traditional medicine because they are economy, have a little side effects and according to W.H.O, about 80% of the world population rely mainly on herbal remedies (Patel *et al.*, 2012). The World Health Organization has recently defined traditional medicine (including herbal drugs) as comprising therapeutic practices that have been in existence, often for hundreds of years, before the development and spread of modern medicine and are still in use today (Agarwal *et al.*, 2012). The uses of traditional medicines are widely spread and plants represent a large source of natural chemicals that might serve as leads for the development of the novel drugs (Gautam *et al.*, 2013). Scientists have devised different ways of alienating the problem and one of the easy and cheapest options is herbal medicines. Herbs have been in use since long time to treat various diseases (Patil *et al.*, 2012). Almost one fourth of pharmaceutical drugs are derived from botanicals (Brown *et al.*, 1959).

Pyrexia or fever is caused as a secondary impact of infection, malignancy or other diseased states. It is the body's natural defense to create an environment where infectious agent or damaged tissue cannot survive (Chattopadhyay *et al.*, 2005). Normally the infected or damaged tissue initiates the enhanced formation of proinflammatory mediator's (Cytokines

like interleukin 1 β , α , β and TNF- α), which increase the synthesis of prostaglandin E2 (PG E2) near peptic hypothalamus area and thereby triggering the hypothalamus to elevate the body temperature (Spacer and Breder 1994). As the temperature regulatory system is governed by a nervous feedback mechanism, so when body temperature becomes very high, it dilate the blood vessels and increasing sweating to reduce the temperature; but when the body temperature become very low hypothalamus protect the internal temperature by vasoconstriction. High fever often increases faster disease progression by increasing tissue catabolism, dehydration and existing complaints, as found in HIV (Veugelers *et al.*, 1997). Drugs having anti-inflammatory activity generally possess antipyretic activity (e.g) non-steroidal anti-inflammatory drugs (NSAIDs). It has been suggested that prostaglandin (PGE) mediates pyrogen fever; the ability of NSAIDs, to inhibit prostaglandin synthesis could help to explain their antipyretic activity.

Fever is one of the most common presenting signs of illness in office-based primary care pediatric practice, accounting for 19% to 30% of visits (Eskerud *et al.*, 1992; Baucher *et al.*, 2001). Infants and young children are particularly susceptible to fever because of their small body size, high ratio of body surface area to weight, and low amount of subcutaneous fat. Although most experts consider fever a beneficial physiologic response to the infectious process, it can lead to patient irritability and stress as well as high parental anxiety (Guton; 1997). Therefore, physicians usually prefer to prescribe antipyretic agents in addition to nonpharmacologic, physical fever-reducing modalities (Baraff., 1993). Pyrexia or Fever is defined as an elevation of body temperature. It is a response due to tissue damage, inflammation, malignancy or graft rejection. Cytokines, interleukin, interferon and Tumor Necrosis Factor α (TNF- α) are formed in large amount under this condition, which increase PGE2 which in turn triggers hypothalamus to elevate body temperature (Rajani *et al.*, 2011). Fever is associated with symptoms of sickness behavior which consist of lethargy, depression, anorexia, sleepiness, & inability to concentrate. This increase in set point triggers increased muscle tone & shivering. However antipyretic medication can be effective at lowering the temperature which may include the affected person's comfort (Duraisankar *et al.*, 2012).

According to Ayurveda, pyrexia originates from a combination of indigestion, seasonal variations and significant alterations in daily routine ^[55]. Due to poor hygiene practices and malnutrition, children in

developing countries frequently suffer from various forms of infections which present as fevers. These fevers are often accompanied by aches and pains which all lead to morbidity and mortality (Gupta et al., 2008).

Fever is a complex physiologic response triggered by infections or aseptic stimuli. Elevation in body temperature occurs when the concentration of prostaglandin E2 (PGE2) increases within parts of the brain. Such an elevation contributes to a considerable alteration in the firing rate of neurons that control the thermoregulation process in the hypothalamus. It is now evident that most of the antipyretic drugs exert their action by inhibiting the enzymatic activity of cyclooxygenase and consequently reducing the levels of PGE2 within the hypothalamic region (Ighodaro et al., 2009).

Solanum virginianum L. commonly known as wild eggplant or nightshade plant, is a prickly herb found in most of the parts of the Asia and Australia of the world. It belongs to family Solanaceae, has spines throughout the plant. Fruits are globular and edible, flowers appear in cymes or sometimes solitary and are blue in colour, leaves are elliptical or ovate and are full of spines, stems appear green when young and brownish when matured.

Solanum virginianum used by the local people as folk medicines in treating throat infections and other inflammatory problems. The fruits are known for several medicinal uses like anthelmintic, antipyretic, laxative, anti-inflammatory, anti-asthmatic and aphrodisiac activities. The fruit paste is applied externally to the affected area for treating pimples and swellings. The various parts of the plant are reputed in indigenous Hindu Medicine to have high medicinal value in various diseases like cough, asthma, fever, heart disease etc.

This study was designed to evaluate different *in vitro* biological activities like antioxidant activity, antimicrobial activity and *in vivo* anti-pyretic and analgesic activities of hydroalcoholic extract of leaves of *Solanum virginianum*.

MATERIAL AND METHODS:

Collection of plant material:

Leaves of *Solanum virginianum* were collected from rural area of Bhopal (M.P), India in the months of January, 2019.

Extraction procedure:

Following procedure was adopted for the preparation of hydroalcoholic extract from the shade dried and powdered herbs (Khandelwal, 2005; Kokate, 1994):

Phytochemical Screening:

The chemical tests were performed for testing different chemical groups present in extracts (Roopashree et al., 2008; Obasi et al., 2010; Audu et al., 2007).

Animals:

Albino Wistar rats of either sex (150–200 g) were group housed (n= 6) under a standard 12 h light/dark cycle and controlled conditions of temperature and humidity (25±2 °C, 55–65%). Rats received standard rodent chow and water *ad libitum*. Animals were acclimatized to laboratory conditions for 7 days before carrying out the experiments. All the experiments were carried in a noise-free room between 08.00 to 15.00 h. Separate group (n=6) of rat was used for each set of experiments. The animal studies were approved by the Institutional Animal Ethics Committee (IAEC), constituted for the purpose of control and supervision of experimental animals by Ministry of Environment and Forests, Government of India, New Delhi, India.

Acute oral toxicity study:

Acute oral toxicity was conducted according to the method of Organisation for Economic Co-operation and Development (OECD) (OECD, 2002). Hydroalcoholic extract of Leaves of *Solanum virginianum* (5, 50, 300, and 2000 mg/kg) was administered orally for 4 days of six groups of rats (n=6) and the animals were kept under observation for mortality as well as any behavioral changes for evaluation of a possible anti-pyretic and analgesic activity.

OECD, 2002. Acute oral toxicity. Acute oral toxic class method guideline 423 adopted 23.03.1996. In: Eleventh Addendum to the, OECD, guidelines for the testing of chemicals organisation for economical co-operation and development, Paris, June, 2000.

Anti-Pyretic activity:

Body weights of the animals were recorded and they were randomly divided into 5 groups of 6 animals each as follows:

Group I served as normal

Group II served as control- animals were treated with yeast via subcutaneous injection (10ml/kg).

Group III animals were administered with yeast (10 ml/kg) and the standard drug paracetamol (150mg/kg b.w.), orally

Group IV animals were administered with yeast (10ml/kg.) and with hydroalcoholic extract of leaves of *Solanum virginianum* (100mg/kg b.w.), orally

Group V animals were administered with yeast (10ml/kg.) and with hydroalcoholic extract of leaves of *Solanum virginianum* (200mg/kg b.w.), orally.

Yeast induced pyrexia:

Pyrexia was induced by subcutaneous injection of 20 % w/v of brewer's yeast (10ml/kg) in distilled water. Basal rectal temperature was measured before the injection of yeast, by inserting digital clinical thermometer to a depth of 2 cm into the rectum. The rise in rectal temperature was recorded 19 h after yeast injection. Paracetamol 150mg/kg body weight was used as the standard antipyretic drug. Rectal temperature of animals was noted at regular intervals following the respective treatments. The temperature was measured at 1st, 2nd, and 3rd hour after drug administration (Niazi et al., 2010).

Statistical analysis:

The values were expressed as mean \pm SEM (n=6). The statistical significance was assessed using one-way analysis of variance (ANOVA) followed by Tukey's test and $P < 0.05$, $P < 0.01$, and $P < 0.001$ were considered to be statistically significant.

RESULTS AND DISCUSSION:

The percentage yield of extraction is very important phenomenon in phytochemical extraction to evaluate the standard extraction efficiency for a particular plant, different parts of same plant or different solvents used. The extractive value of petroleum ether and hydroalcoholic extract of *Tinospora sinensis* was found 1.69 and 7.45 respectively. The Hydroalcoholic extract of leaves of *Solanum virginianum* had revealed the presence of flavonoids, Phenols, carbohydrates, Alkaloids and saponins. Protein and diterpenes were found to be absent.

The content of total phenolic compounds (TPC) content was expressed as mg/100mg of gallic acid equivalent of dry extract sample using the equation obtained from the calibration curve: $Y = 0.042X - 0.002$, $R^2 = 0.999$, where X is the gallic acid equivalent (GAE) and Y is the absorbance.

The content of total flavonoid compounds (TFC) content was expressed as mg/100mg of quercetin equivalent of dry extract sample using the equation obtained from the calibration curve: $Y = 0.06X + 0.019$, $R^2 = 0.999$, where X is the quercetin equivalent (QE) and Y is the absorbance. Total phenolic and total flavonoid content of

hydroalcoholic extract of *Solanum virginianum* was found 1.024 and 1.358 respectively.

This study was carried out to evaluate the antipyretic effects of hydroalcoholic extract of leaves of *Solanum virginianum* in rats. The antipyretic potential was done using yeast as pyrexia inducing agent. The findings from the present study were in agreement with other studies on antipyretic potential medicinal plants in animal models. Similar work by demonstrated antipyretic effects of aqueous and methanolic root extracts of *Asparagus racemosus* in yeast-induced pyrexia. Besides, similar findings were reported by on the antipyretic effect of *Urtica dioica* (L.) aqueous leaf extract against brewer's yeast-induced fever in animal models. A study by demonstrated the antipyretic effects of selected medicinal plants in albino rats (Nagaveni et al., 2011).

The antipyretic effects of hydroalcoholic extract of leaves of *Solanum virginianum* at different dose levels exhibited a dose-independent response on yeast-induced fever in rats. Lower dosages of 100 mg/kg and higher 200 mg/kg body weight were as effective as dose dependent manner. The results of present study indicate that the hydroalcoholic extract of *Solanum virginianum* leaves possesses significant antipyretic effect on yeast induced hyperthermia in rats. This may be attributed to the presence of chemical constituents in the extracts which may be involved in inhibition of prostaglandin synthesis. Also, there are several mediators or multiprocessors underlining the pathogenesis of fever. Inhibition of any of these mediators may bring about antipyresis. The hydroalcoholic extract of *Solanum virginianum* leaves at the 200 mg/kg body weight was marginally effective than 100 mg/kg body weight dose. These findings suggest a better or a similar prostaglandin synthesis inhibition by the active components in the plant's extracts. There is therefore, a possibility of the plant extracts working effectively by blocking alternative mechanisms during fever inhibition.

The tail immersion assay is a thermal test for evaluating the analgesic potential of compounds. Tail immersion responses are believed to be spinally mediated reflex. Analgesics are drugs used to treat or reduce pain and the classical analgesic drugs notably opiates and non-steroidal anti-inflammatory drugs have their origin in natural products but many synthetic compounds that act by the same mechanism have been developed and are associated with serious adverse effects such as ulceration, gastrointestinal bleeding, additive potential, respiratory distress, drowsiness, nausea etc (Laurence et al., 1997; Mate

et al., 2008). In the tail immersion, hydroalcoholic extract of *Solanum virginianum* leaves administered at a dose of 100 and 200 mg/kg p.o has shown

analgesic effect with reaction time at 0, 30, 60 and 120 min, respectively.

Table 1: % Yield of hydroalcoholic extract of leaves of *Solanum virginianum*

S. No.	Extracts	% Yield (w/w)
1.	Petroleum ether	1.69
2.	Hydroalcoholic	7.45

Table 2: Phytochemical screening of leaves of *Solanum virginianum*

S. No.	Constituents	Hydroalcoholic extract
1.	Alkaloids Dragendroff's test Hager's test	-ve +ve
3.	Flavonoids Lead acetate Alkaline test	+ve +ve
4.	Phenolics FeCl ₃	+ve
5.	Proteins Xanthoproteic test	-ve
6.	Carbohydrates Fehling's test	+ve
7.	Saponins Foam test	+ve
8.	Diterpenes Copper acetate test	-ve

Table 3: Total phenolic and total flavonoid content of extract of *Solanum virginianum*

S. No.	Extract	Total Phenol (GAE) (mg/100mg)	Total flavonoid (QE) (mg/100mg)
1.	Hydroalcoholic extract	1.024	1.358

Table 4: Antipyretic activity of hydroalcoholic extract of leaves of *Solanum virginianum* against yeast induced pyrexia in rats

Rectal Temperature in °C after 18hrs of Yeast Injection				
Group	0 hr	1 hr	2 hr	3 hr
Group I	38.40±0.51	38.10±0.52	38.00±0.50	37.50±0.51
Group II	42.50±0.15	42.00±0.15	41.50±0.15	41.20±0.15
Group III	42.50±0.50	39.50±0.50	38.50±0.51*	37.50±0.51*
Group IV	42.00±0.50	41.50±0.50	40.50±0.50	39.50±0.51*
Group V	42.00±0.50	41.00±0.50	39.30±0.51*	38.60±0.50*

Values expressed as mean ± SEM (n=6) *P<0.05 as compared to pyretic control

Table 5: Effect of hydroalcoholic extract of leaves of *Solanum virginianum* and Indomethacin as compared to control group at different time intervals tail immersion method

Groups	Dose of extract (mg/kg) p.o.	Reaction time in seconds			
		0 min	30 min	60 min	120min
Control (0.1 ml of 1% w/v)	-	1.40± 0.20	1.50± 0.20	1.60± 0.20	1.65± 0.20
Indomethacin	10	3.50± 0.10	6.00± 0.10	7.50± 0.10**	7.80± 0.10***
Hydroalcoholic extract of <i>Solanum virginianum</i> leaves	100	2.10± 0.13	5.20± 0.14	5.30± 0.18*	5.80± 0.15*
Hydroalcoholic extract of <i>Solanum virginianum</i> leaves	200	3.30± 0.15	6.10± 0.16	7.00± 0.10**	7.10± 0.16***

All values are expressed as mean ± SD; *P < 0.05 v/s control

CONCLUSION:

In conclusion, this study has revealed that these plants are endowed the many bioactive compounds which possess antipyretic and analgesic activity in rats. The effect of hydroalcoholic extract of leaves of *Solanum virginianum* were determined after administration at two dose levels (100 and 200 mg/kg b.w.) in yeast induced pyrexia and tail immersion in rats. Our findings suggest a medicinal plant with multiple therapeutic properties that could serve as an antipyretic drug. To elucidate the exact mechanism of action of these active constituents, further investigation is required.

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