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Antinociceptive and Antipyretic Effects of *Strychnos potatorum* Linn. Seeds on Experimental Rats

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Abstract: In traditional system of medicine, *Strychnos potatorum* Linn. seeds were used for various ailments like pain, inflammation, anemia, jaundice, bronchitis, diabetes, conjunctivitis, chronic diarrhoea, dysentery etc. To investigate the folkloric use, the present study was carried out to evaluate the antinociceptive and antipyretic activities of seed powder (SPP) and aqueous extract (SPE) of *Strychnos potatorum* Linn seeds in experimental wistar albino mice and rats, respectively. The antinociceptive activity was studied in both chemical [acetic acid (1 mL/100 g b.wt.) induced writhing] and thermal (hot plate and tail immersion technique) models of inducing nociception. Administration of SPP and SPE at two dose levels (100 and 200 mg kg^{-1} p.o.) significantly ($p<0.001$) decreased the abdominal contractions in acetic acid induced writhing model and significantly ($p<0.001$) increased the reaction time in both hot plate and tail immersion techniques, when compared with the standard drug Aspirin (100 mg kg^{-1}, p.o.). Thus SPP and SPE were found to exhibit antinociceptive activity in both chemical and thermal models indicating their central as well as peripheral mechanisms in inhibiting the nociception, respectively. The antipyretic activity was studied by injecting TAB vaccine at the dose of 1 mL kg^{-1} b.wt., where the pyresis was induced after 6 h. Both SPP and SPE exhibited dose dependent activity in reducing the pyrexia which is comparable to that of Paracetamol (100 mg kg^{-1}, p.o.).

Key words: *Strychnos potatorum*, acetic acid induced writhing, hot plate method, tail immersion, TAB vaccine induced pyrexia

INTRODUCTION

The medicinal use of herbs is widespread. Plants synthesize complex (organic) molecules for their structure and function and therefore are a rich source of chemicals. Many herbs have been shown to possess significant anti-inflammatory properties. Along with being used as an anti-inflammatory, some herbs have the potential to be used as analgesics as well. Herbs in various forms have been used to treat joint pains for centuries. Much of the testing of herbal remedies is still in the early stages and it is difficult to assess the true effectiveness of these herbs.

Strychnos potatorum Linn. (Fam: Loganiaceae) is a moderate sized tree found in southern and central parts of India, Srilanka and Burma (Kirtikar and Basu, 1933). Selected parts of the tree like seeds, ripe fruit and roots are used in traditional system of medicine for the treatment of various ailments like tumors, pain, inflammation, anemia and jaundice (Kirtikar and Basu, 2000). In English, it is commonly known as clearing nut tree. The ripe seeds are used for clearing muddy water. They are reported to be very effective as coagulant aids. It is effective in removing the suspended impurities. The clarification is due to the combined action of colloids and alkaloids in the seeds. The albumin and other colloids sensitize the suspension and the coagulation is then caused by the alkaloidal ions (Wealth of India, 1967).

The seeds were reported to have various phytochemicals like alkaloids-diaboline and its acetate (Singh and Kapoor, 1980), brucine, loganin, mannose, sucrose, arachidonic, lignoceric, linoleic, oleic, palmitic and stearic acids (Singh and Bajpai, 1975) steroids and triterpenes (Singh *et al*., 1975a, b), polysaccharides (mannogalactans) (Rao *et al*., 1991; Corsaro *et al*., 1995) etc. Mallikarjunah *et al*. (2007) studied the detailed phytochemical analysis of *Strychnos potatorum* root, stem bark and seeds which revealed the presence of alkaloids, flavonoids, saponins and other phenols. The HPLC profile of the alkaloids present was also reported. Sanmugapriya and Venkataraman (2010) studied the

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detailed pharmacognostical and phytochemical parameters of the seed, where the secondary metabolites were identified by HPTLC.

Various pharmacological activities like antidiarrhoeal (Swati *et al*., 2002), CNS effects (Singh and Kapoor, 1980), antidiabetic (Mathuram *et al*., 1981) and diuretic (Biswas *et al*., 2001) activities have also been reported. In earlier studies, the seeds were reported to have anti-inflammatory (Sanmugapriya and Venkataraman, 2007a), antiulcerogenic (Sanmugapriya and Venkataraman, 2006a) and hepatoprotective (Sanmugapriya and Venkataraman, 2006b) activities. The acute and chronic toxicity studies of the seeds were also reported Sanmugapriya and Venkataraman, 2007b) which revealed that SPP and SPE were found to be nontoxic up to the dose level of 2000 mg kg^{-1}, p.o. The alkaloids isolated from *Strychnos nuxvomica* (another species) were reported to have significant analgesic and anti-inflammatory activities (Yin *et al*., 2003). Taking consideration of the folkloric use and the anti-inflammatory activity of the seeds reported, the present study was designed to evaluate the antinociceptive and antipyretic properties of *Strychnos potatorum* Linn. seeds in chemical and thermal models of nociception and TAB vaccine induced pyrexia, respectively.

MATERIALS AND METHODS

Plant material: The seed specimens used for the study were collected from crude drug market, Chennai in the year 2002. The identity and genuinity of the seed specimen was confirmed by Dr. S. Jayaraman, Botanist, Plant Anatomy Research Centre, Chennai, Tamilnadu. A voucher specimen has been deposited in the Department of Pharmacology and Environmental Toxicology, University of Madras.

Animals: Wistar albino mice (20-25 g) and rats of either sex (140±20 g) procured from TANUVAS (Tamilnadu University of Veterinary and Animal Sciences), 6 animals per group were used for the study. The animals were kept in polypropylene cages and maintained at a temperature of 22±2°C. They were fed with standard pelleted feed (TANUVAS) and water *ad libitum*. The study has got approval from the Institutional Animal Ethical Committee (IAEC).

Preparation of the extract: The air-dried seeds were coarsely powdered and subjected to hot water decoction for 2 h at 100°C, it was then filtered through muslin cloth and the filtrate was evaporated to dryness. A grey colored semisolid mass was obtained which was dried under vacuum and kept in a dessicator. The percentage yield of the extract (SPE) was 22.5% w/w from the starting crude material. The seed powder (SPP) was prepared by grinding the dried seeds in a blender and used for the study. For the experiment, both the drugs at the doses of 100 and 200 mg kg^{-1}, p.o., respectively {SPP (I and II) and SPE (I and II)} were triturated with distilled water and administered orally immediately.

Phytochemical analysis: SPP and SPE were subjected to phytochemical screening through qualitative chemical analysis and HPTLC fingerprinting.

Antinociceptive activity

Chemical method: Acetic acid induced writhing test: The control group received 1%v/v (1 mL/100 g b.wt.) of acetic acid solution intraperitoneally and from the onset of wriths, the total number of writhing responses was counted for a period of 10 min. The other groups received the respective drugs and after 1 h acetic acid was administered. The onset and the severity of writhing response were noted (Koster *et al*., 1959).

Thermal method

Hot plate test: The basal reaction time for thermal stimulus was observed in the animals by placing on a hotplate maintained at constant temperature (55°C). The drugs were administered and the reaction time of the animals was recorded after 1 h. A cut off period of 15 secs was observed to avoid damage to the paws (Eddy and Leimbach, 1953).

Tail immersion technique: A constant temperature of 50±0.2°C was maintained in a water bath, in which the terminal 3 cm of animal tail was immersed. While nociception measurements were being made, the animals were briefly immobilized (25-30 sec) by using a commercial restrainer. Baseline latencies were determined twice, 5 min apart and averaged to give a single predrug latency. The respective drugs were administered orally and after 1 h, the tail withdrawal latencies were measured. In order to minimize the tissue injury due to repeated exposure of the heat stimulus, a cut off time of 15 sec was imposed. Aspirin (100 mg kg^{-1}, p.o.) is used as the standard drug for comparison (Statile *et al*., 1988).

Antipyretic activity: Wistar albino rats of either sex weighing 140±20 g were selected for the study. Animals were fasted overnight allowing water *ad libitum*. Initial rectal temperatures were recorded using clinical thermometer. Hyperthermia was induced in rats according to the method of Pendse *et al*. (1977) by subcutaneous

injection of 1 mL kg^{-1} b.wt. of TAB vaccine. When the maximum temperature was attained, i.e., after 6 h of TAB vaccine injection, rats which developed satisfactory pyrexia (1°C or more increase in rectal temperature) were only used. Test drugs were administered and the rectal temperature of animals was recorded at hourly intervals for 4 h.

Statistical analysis: The data represents Mean±SEM. Results were analysed statistically using one-way ANOVA followed by Tukey's multiple comparison. The minimum level of significance was set at $p<0.05$.

RESULTS AND DISCUSSION

In acetic acid induced writhing test, both drugs SPP (I and II) and SPE (I and II) exhibited significant ($p<0.001$) analgesic activity, when compared with the control group, whereas [SPE-II (200 mg kg^{-1}, p.o.)] possessed greater activity by showing 51.45% of protection which is comparable with the standard drug, Aspirin (51.86%) (Fig. 1).

In thermal models like hot plate and tail immersion tests, SPP and SPE showed significant ($p<0.001$) analgesic activity by increasing the reaction time (latency time) when compared with the control (Fig. 2, 3). In hot plate test, the Pain Inhibition Percentage (PIP) values showed that the drugs possess analgesic activity in a dose dependent manner and SPE-II (200 mg kg^{-1}, p.o.) exhibited significant analgesic activity by showing 71.87% of PIP. In tail immersion test, the drugs showed greater activity after 60 min of administration by showing increased percentage maximum possible effect (MPE %), in which SPE-II (200 mg kg^{-1}, p.o.) again showed higher activity by exhibiting 64.07% MPE.

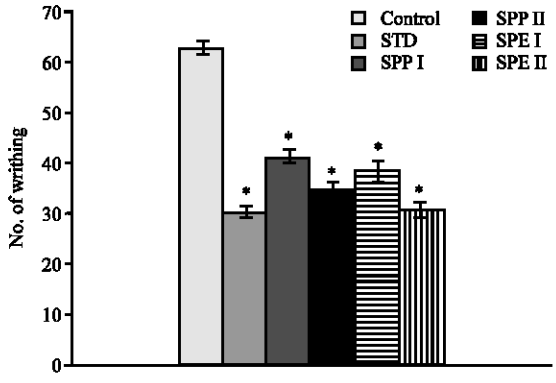

Fig. 1: Effect of SPP and SPE on acetic acid induced writhing in mice. Data represents Mean±SEM of 6 animals. *$p<0.001$ compared to control (One way ANOVA followed by Tukey's multiple comparison test)

SPP (I and II) and SPE (I and II) showed to possess antipyretic activity, which is significant when compared with the control group (Fig. 4). The response in higher dose (200 mg kg^{-1}, p.o.) ($p<0.001$) of the drugs was almost comparable to that of paracetamol (100 mg kg^{-1}, p.o.) ($p<0.001$), the standard drug used for comparison.

The abdominal writhing induced by acetic acid involves the process of the release of Arachidonic Acid (AA) metabolites via cyclooxygenase (COX) and PG biosynthesis (Elisabetsky *et al*., 1995). Several other mediators like kinins, substance P, acetylcholine, are also involved in visceral pain nociception (Jain *et al*., 1997). Thus, the observed effects of both the drugs SPP and SPE suggests the inhibition of prostaglandins and other mediators involved in nociception. Although, the abdominal constriction response induced by acetic acid

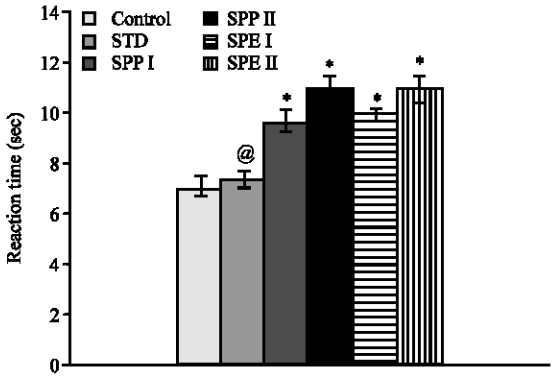

Fig. 2: Effect of SPP and SPE on hot plate test. Data represents Mean±SEM of 6 animals. *$p<0.001$, @$p<0.05$ compared to control (One way ANOVA followed by Tukey's multiple comparison test)

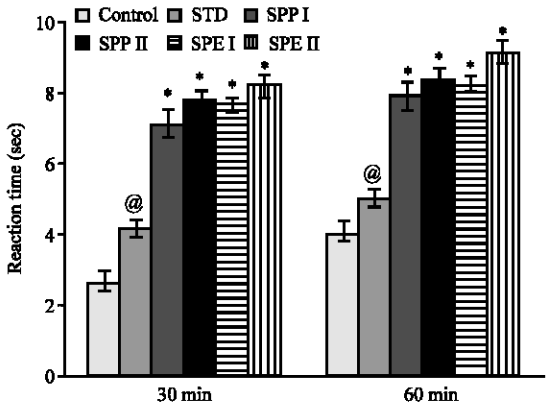

Fig. 3: Effect of SPP and SPE on tail immersion test. Data represents Mean±SEM of 6 animals. *$p<0.001$, @$p<0.05$ compared to control (One way ANOVA followed by Tukey's multiplecomparison test)

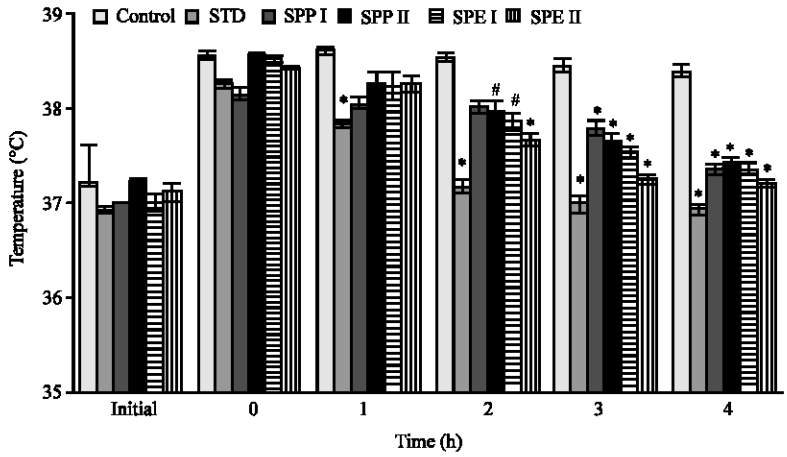

Fig. 4: Effect of SPP and SPE on rectal temperature increase in TABvaccine induced pyrexia. Data represents Mean±SEM of 6 animals. *$p<0.001$, #$p<0.01$ compared to control (One way ANOVA followed by Tukey's multiple comparison test)

is a very sensitive procedure that enables the detection of both central and peripheral antinociceptive activity of compounds in laboratory animals (Sarkhail *et al*., 2003), it is not a very selective pain test, as it shows false positives occurring with sedatives, muscle relaxants and other pharmacological activities (Elisabetsky *et al*., 1995). Due to this lack of specificity, caution is required in interpreting the results until other tests have been performed. Analgesic effects of morphine and other narcotic analgesics (central analgesics) are easily evaluated using thermal nociceptive tests as these tests are more sensitive to opioid μ agonists (Tjolsen *et al*., 1991; Aydin *et al*., 1999). In support of this, the thermal models of nociception are studied to conclude the mechanism of action. In the thermal models also, the SPP and SPE were found to show significant antinociceptive activity. Thus SPP and SPE were found to exhibit antinociceptive activity in both chemical and thermal models indicating their central as well as peripheral mechanisms in inhibiting the nociception, respectively.

It is well known that the antipyretic agents act by inhibiting prostaglandin synthesis, since PG production in the CNS is the final common pathway responsible for fever induction (Panthong *et al*., 2003). Similarly, it is logical to presume that SPP and SPE may also exert its effects through the same mechanism, since their analgesic activity was also presumed to be mediated through PG inhibition.

From previous reports by Mallikharjunah *et al*. (2007) and in the preliminary phytochemical and HPTLC screening (Sanmugapriya and Venkataraman, 2010), both SPP and SPE revealed the presence of steroids, triterpenoids, saponins and polyphenolics. The presence of these phytochemicals may be responsible for the analgesic activity of the drugs SPP and SPE.

CONCLUSION

In conclusion, the drugs (SPP and SPE) showed both central and peripheral analgesic activities in a dose dependent manner; among which SPE-II (200 mg kg^{-1}, p.o.) possessed significant ($p<0.001$) activity, when compared with the standard drug, Aspirin (100 mg kg^{-1}, p.o.). It also shown to have significant dose dependent antipyretic activity in TAB vaccine induced pyrexia model.

REFERENCES

Aydin, S., T. Demir, Y. Ozturk and K.H.C. Baser, 1999. Analgesic activity of *Nepeta italica* L. Phytother. Res., 13: 20-23.

Biswas, S., T. Murugesan, K. Maiti, L. Ghosh, M. Pal and B.P. Saha, 2001. Study on the diuretic activity of *Strychnos potatorum* Linn, seed extract in albino rats. Phytomedicine, 8: 469-471.

Corsaro, M.M., I. Giuddiciani, R. Lanzetta, C.E. Marciano, P. Monaco and M. Parilli, 1995. Polysaccharides from seeds of *Strychnos* species. Phytochemistry, 39: 1377-1380.

Eddy, N.B. and D. Leimbach, 1953. Synthetic analgesics II. Diethienylbutenyl and diethynybutlylamines. J. Pharmacol. Exp. Ther., 107: 385-393.

Elisabetsky, E., T.A. Amador, R.R. Albuquerque, D.S. Nunes and A.C.T. Cavalho, 1995. Analgesic activity of psychotria colorata (Wild ex R and S). muell arg. alkaloids. J. Ethnopharmacol., 48: 77-83.

Jain, S.C., R. Jain, R.A. Sharma and F. Capasso, 1997. Pharmacological investigation of *Cassiaitalica*. J. Ethnopharmacol., 58: 135-142.

Kirtikar, K.R. and B.D. Basu, 1933. Indian Medicinal Plants. Vol. I, Lalit Mohan Basu and Co., Allahabad, pp: 100-108.

Kirtikar, K.R. and B.D. Basu, 2000. Illustrated Indian Medicinal Plants. Sir Satguru's Publications, Delhi, pp: 2271.

Koster, R., M. Anderson and E.J. De Beer, 1959. Acetic acid for analgesic screening. Fed. Proc., 18: 412-418.

Mallikharjunah, P.B., L.N. Rajanna, Y.N. Seetharam and G.K. Sharanabasappa, 2007. Phytochemical studies of *Strychnos potatorum* L.f.-A medicinal plant. E-J. Chem., 4: 510-518.

Mathuram, L.N., H.C. Samanna, V.M. Ramasamy and R. Natarajan, 1981. Studies on the hypoglycemic effects of *Strychnos potatorum* and *Acacia arabica* on alloxan diabetes in rabbits. Cheiron, 10: 1-5.

Panthong, A., D. Kanjanapothi, T. Taestikul, T. Wongcome and V. Reutrakul, 2003. Anti-inflammatory and antipyretic properties of *Clerodendron petasites* S. Moore. J. Ethnopharmacol., 85: 151-156.

Pendse, V.K., A.P. Dadich, P.N. Mathur, M.S. Bal and B.R. Madan, 1977. Antinflammatory, Immunosuppressive and some related pharmacological actions of the water extract of Neem Giloe (*Tinospora cordifolia*): A preliminary report. Ind. J. Pharmacol., 9: 221-224.

Rao, E.V., K.S. Ramana and M. Venkateswarao, 1991. Revised structure and antihypercholesterolemic activity of a mannogalactan from *Strychnos potatorum*. Ind. J. Pharm. Sci., 53: 53-57.

Sanmugapriya, E. and S. Venkataraman, 2006a. Studies on hepatoprotective and antioxidant actions of *Strychnos potatorum* Linn seeds on CCl_4-induced acute hepatic injury in experimental rats. J. Ethnopharmacol., 105: 154-160.

Sanmugapriya, E. and S. Venkataraman, 2006b. Toxicological investigations on *Strychnos potatorum* Lin seeds in experimental animal models. J. Health Sci., 52: 339-343.

Sanmugapriya, E. and S. Venkataraman, 2007a. Antiulcerogenic potential of *Strychnos potatorum* Lin seeds in aspirin+pylorus ligation induced ulcers in experimental rats. Phytomedicin, 14: 360-365.

Sanmugapriya, E. and S. Venkataraman, 2007b. Antiinflammatory effect of *Strychnos potatorum* seeds on acute and sub acute inflammation in experimental rat models. Pharm. Biol., 45: 435-439.

Sanmugapriya, E. and S. Venkataraman, 2010. Pharmacognostical and phytochemical studies of *Strychnos potatorum* Linn seeds. PHCOG. J., 2: 190-197.

Sarkhail, P., M. Abdollahi and A. Shafiee, 2003. Antinociceptive effect of *Phlomis olivieri* Benth., *Phlomis anisodonta* Boiss. and *Phlomis persica* Boiss. total extracts. Pharmacol. Res., 48: 263-266.

Singh, A. and R.K. Bajpai, 1975. Chemical examination of fixed oil from the seeds of *Strychnos potatorum*. Linn F. mixed fatty acids. J. Ind. Chem. Soc., 3: 768-769.

Singh, H., V.K Kapoor, J.D. Phillipson and N.G. Bisset, 1975a. Diaboline from *Strychnos potatorum*. Phytochemistry, 14: 587-588.

Singh, H., V.K. Kapoor and M.S. Manhas, 1975b. Investigation of *Strychnos* sp. III. Study of triterpenes and sterol of *Strychnos potatorum* seeds. Planta Med., 28: 392-396.

Singh, H. and V.K. Kapoor, 1980. Pharmacological studies of alkaloids of *Strychnos potatorum* seeds. Planta Med., 38: 133-137.

Statile, L., M.M. Puig, W. Warner, M. Bansinath, M. Lovitz and H. Turndorf, 1988. Droperidol enhances fentanyl and sufentanil but not morphine analgesia. Gen. Pharmacol.: Vascular Syst., 19: 451-454.

Swati, B., T. Murugesan, S. Sinha, K. Maiti, J.R. Gayen, M. Pal and B.P. Saha, 2002. Antidiarrhoeal activity of *Strychnos potatorum* seed extract in rats. Fitoterapia, 73: 43-47.

Tjolsen, A., J.H. Rosland, O.G. Berge and K. Hole, 1991. The increasing temperature hot-plate test: An improved test of nociception in mice and rats. J. Pharmacol. Methods, 25: 241-250.

Wealth of India, 1967. Raw Materials. Vol. 10, Publications and Information Directorate, CSIR., New Delhi, pp: 66-67.

Yin, W., T.S. Wang, F.Z. Yin and B.C. Cai, 2003. Analgesic and anti-inflammatory properties of brucine and brucine N-oxide extracted from seeds of *Strychnos nux-vomica*. J. Ethnopharmacol., 88: 205-214.