Screening of anti-pyretic potential of various parts of *Pterospermum canescens*, Roxb., (Sterculiaceae) extracts in experimental animals.

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Abstract: Pyrexia is one of the most common symptomatic presentations of disease. A lot of research is going on worldwide towards finding antipyretic agents from the natural sources. The main aim of the present study was to evaluate the antipyretic potential of petroleum ether, chloroform and methanol extracts (100 mg/kg, 200 mg/kg) of *Pterospermum canescens*, Roxb., (Sterculiaceae (leaves, stem and stem bark) was investigated for its antipyretic activity. Pyrexia was induced in Wistar Albino rats by Brewer’s yeast (10mg/kg), were used in this study to assess anti-pyretic potential of the plant using Indomethacin as standard (10 µg/kg). Petroleum ether, chloroform and methanol extracts of leaf, stem and stem bark were exhibited (P < 0.001) anti-inflammatory activity at 100 mg/kg and 200 mg/kg doses when compared with the standard, while methanol stem (100mg/kg) extract exhibited significant activity (p <0.05).

Key words: *Pterospermum canescens*, Antipyretic activity, Brewer’s yeast, Thermometer, Indomethacin.

Introduction

Plants are important and basic of preventive and curative healthcare system since immemorial. Disease is as old as mankind and use of indigenous herbal medicine is a very ancient art and an integral part of treatment¹. According to WHO, nearly, 75-80% of world population still depends on herbal medicines. Active constituents from plant sources directly used as therapeutic agent and phytoconstituents are also served as lead molecule for the synthesis of various drugs². WHO noted that about 25% of modern medicines are descended from plant sources used traditionally and research on traditional medicinal herbal plant leads to discovery of 75% herbal drugs³.

Pyresis is a clinical condition that results in increase in body temperature. 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

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Normal body temperature is regulated by a center in the hypothalamus that ensures a balance between heat and loss and production. Fever occurs when there is a disturbance of this hypothalamic ‘thermostat’, which leads to the set point of body temperature being raised. Once there has been a return to the normal set point, the temperature regulating mechanisms (dilatation of superficial blood vessels, sweating, etc.) then operate to reduce the elevated body temperature. Normally the infected or damaged tissue initiates the enhanced formation of proinflammatory mediator’s (Cytokines like interleukin 1β, 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. 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. High fever enhances faster disease progression by increasing tissue catabolism, dehydration and existing complaints. 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. Search for safe herbal remedies with potent antipyretic activity received momentum recently as the available antipyretics, such as paracetamol, aspirin, nimusulide etc, which have toxic effect to the various organs of the body.

With this background, this study was conducted with an objective of evaluation of the antipyretic activity of *Pterospermum canescens*, Roxb., in Wistar albino rats. The genus *Pterospermum* Schreb., (Sterculiaceae) represents of about 40 species in the world, of which 12 species were reported from India and 8 species has been reported from Tamil Nadu state and is also available in the dry evergreen forests of Sri Harikotta Island, Nellore District, Andrapradesh as well as in Coramantal coast.

An ethnomedicinal plant species *Pterospermum canescens* Roxb., (Syn. *Pterospermum suberifolium* Lam.) locally known as *Sempulavu* was distributed in all districts of Tamil Nadu. Ethnomedicinally, the leaves are used for headache, treatment of fractured bones, small pox, antimicrobial and anti-inflammatory properties. The plant has been reported to contain β- amyrin, betulin, kaempferol, lupeol, quercetin, scopoletin and β - sitosterol and α-sitosterol, 3, 7, 11, 15- tetramethyl-2-hexa decane-1-ol, ricinoleic acid, vitamin-E, phytol, α-tocopherol, diethyl phthalate, squalene, benzhydrazide-3-mthoxy-N2-(4-henylcyclo hexylideno, benzoic acid, 4- heptyl-4-cyanophenyl ester and n-hexadecanoic acid. After the scrutiny of literatures, it was confirmed that so far no other work has been carried out on this plant. Hence, the present study aims to develop an antimicrobial lead of therapeutic interest from this selected ethnomedicinal plant.

Materials And Methods

Plant material

The plant material (leaf, stem and stem bark) of *Pterospermum canescens* Roxb., were collected from the Kallapet vicinity of Pondicherry and the collected plant material was botanically identified and confirmed by the Plant Taxonomist Dr. A.C. Tangavelou and the herbarium specimen (KPJ 42) was prepared and deposited at the department for future reference. The leaves, stem as well as stem bark were separately dried in shade.

Preparation of extracts

The collected plant material (leaf, stem, stem bark) were chopped into small pieces, shade dried and coarsely powdered by using a pulverizer and then pass it through a 40 mesh size sieve. Then, the powder materials were subjected to successive solvent extraction with organic solvents of increasing polarity such as petroleum ether, chloroform and methanol by continuous hot percolation method using soxhlet apparatus. The solvents were then evaporated to dryness under reduced pressure in a Rotary Evaporator at 40- 45°C. The concentrated extracts of leaves, stem and stem bark were separately aliquoted in amber-coloured bottles and kept in desiccator for further use. The resulted extracts were used for screening of antipyretic activity.
Animals

Wistar albino rats (180 – 230 g) were used for the pharmacological studies. They were kept in polypropylene cages at 25 ± 2°C, with relative humidity 45-55% under 12 h light and dark cycles. All the animals were acclimatized to the laboratory conditions for a week before use. They were fed with standard animal feed (Kamadhenu agencies, Bangalore, India) and water *ad libitum*. The experimental protocols were carried out at C.L. Baid Metha College of Pharmacy, Thoarpakkam, Chennai (IAEC/ 34/ 22/ CLBMCP/ 2011, dated on 7/2/2011) approved by the Institutional Animal Ethics Committee.

Antipyretic activity Induction of pyrexia

Induction of pyrexia

Wistar albino rats were used for the screening of antipyretic activity. Briefly, pyrexia was induced in the animals that have been deprived of feeds for 6 h but were adequately supplied with water *ad libitum* by subcutaneous administration of 20% w/v of brewer’s yeast at a dose of 10 mg/kg body weight to near the groin of the animals. The rectal temperature of the rats were measured 17 h after brewer’s yeast injection by inserting the digital thermometer, 3-4 cm into the rectum and only rats that showed an increase of at least 0.5°C rise in temperature were used only for the study.

Animal grouping and administration of extracts

Antipyretic activity of *Pterospermum canescens* Roxb., was carried out by Brewer’s yeast induced pyrexia method. The rats were divided into eight groups of six animals each, respectively at doses of 100 and 200 mg/kg body weight. The dose of the extracts was selected on the basis of folkloric use of the plant. Group I served as control (0.9% Normal saline with 3% Tween, 2 ml/kg), Group II, III (PETL, PETH - 100, 200 mg/kg); Group IV, V (CHL, CHH - 100, 200 mg/kg) and Group VI, VII (MEL, MEH - 100, 200 mg/kg) are administered with petroleum ether, chloroform and methanol extracts of *Pterospermum canescens*, Roxb., (leaf, stem, stem bark) respectively and Group VIII served as standard (Indomethacin, 10 mg/kg, orally). All groups of animals were administered with their doses (17 h after induction of pyrexia) and brewer’s yeast preparation by using plastic syringes attached to a metal oropharyngeal cannula. The animals were allowed free access to rat pellets and tap water after their daily doses. The rectal temperature was measured at 0, 30, 60, 120 and 180 minutes after their doses.

Statistical analysis

Data were expressed as mean ± SEM and were analyzed statistically by one way ANOVA procedures; followed by using Dunnett’s test. A difference was considered significant as $P < 0.05$.

Results and Discussions

Antipyretic activity was evaluated by Brewer’s yeast induced pyrexia in rats and the rectal temperature was reduced at 0, 30, 60, 120 and 180 minutes by insertion of a clinical thermometer to a depth of 2 cm into rectum.

### Table No.1: Antipyretic activity of leaf extracts

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Rectal temperature in °C</th>
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<tr>
<td>Control</td>
<td>36.78 ± 0.13</td>
</tr>
<tr>
<td>PETL</td>
<td>36.98 ± 0.14</td>
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<tr>
<td>PETH</td>
<td>35.65 ± 0.20</td>
</tr>
<tr>
<td>CHL</td>
<td>36.05 ± 0.24</td>
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<tr>
<td>CHH</td>
<td>36.13 ± 0.28</td>
</tr>
<tr>
<td>MEL</td>
<td>35.13 ± 0.07</td>
</tr>
<tr>
<td>MEH</td>
<td>35.10 ± 0.04</td>
</tr>
<tr>
<td>STD</td>
<td>35.37 ± 0.11</td>
</tr>
</tbody>
</table>

Values shown are mean ± SEM (n= 6). $^{*}P < 0.01$, $^{*}P < 0.05$ experimental groups were compared with control.
Figure 1. Antipyretic activity of leaf extracts

Table No. 2: Antipyretic activity of stem extracts

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Rectal temperature in °C</th>
<th>0 min</th>
<th>30 min</th>
<th>60 min</th>
<th>120 min</th>
<th>180 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>36.78 ± 0.13</td>
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<td>36.78 ± 0.13</td>
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<tr>
<td>PETL</td>
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<tr>
<td>PETH</td>
<td>35.87 ± 0.13</td>
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<tr>
<td>CHL</td>
<td>35.93 ± 0.23</td>
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<tr>
<td>CHH</td>
<td>36.02 ± 0.24</td>
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<tr>
<td>MEL</td>
<td>35.43 ± 0.14*</td>
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<tr>
<td>MEH</td>
<td>35.32 ± 0.10</td>
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<tr>
<td>STD</td>
<td>35.37 ± 0.11</td>
<td>35.37 ± 0.11</td>
<td>35.37 ± 0.11</td>
<td>35.37 ± 0.11</td>
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</tbody>
</table>

Values shown are mean ± SEM (n = 6). *P < 0.01, **P < 0.05 experimental groups were compared with control.
Table No.3: Antipyretic activity of stem bark extracts

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Rectal temperature in °C</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0 min</td>
</tr>
<tr>
<td>Control</td>
<td>36.78 ± 0.13</td>
</tr>
<tr>
<td>PETL</td>
<td>36.83 ± 0.15</td>
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<tr>
<td>PETH</td>
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<tr>
<td>CHL</td>
<td>35.93 ± 0.23</td>
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<tr>
<td>CHH</td>
<td>36.02 ± 0.24</td>
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<tr>
<td>MEL</td>
<td>35.43 ± 0.14</td>
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<tr>
<td>MEH</td>
<td>35.32 ± 0.10</td>
</tr>
<tr>
<td>STD</td>
<td>35.37 ± 0.11</td>
</tr>
</tbody>
</table>

Values shown are mean ± SEM (n= 6). **P < 0.01, *P < 0.05 experimental groups were compared with control.

Figure 3. Antipyretic activity of stem bark extracts

Leaf

In Brewer’s yeast induced pyrexia method, standard group of animals were exhibited antipyretic activity when compared with the control group of animals. Petroleum ether, chloroform and methanol (100, 200 mg/kg) leaf extracts didn’t show significant, but exhibited antipyretic activity when compared with the control group of animals (Fig. 1; Table No.1).

Stem

In Brewer’s yeast induced pyrexia method, standard group of animals were exhibited antipyretic activity when compared with the control group of animals. Methanol (100 mg/kg) stem extract was exhibited significant (P < 0.05), antipyretic activity when compared with the control group of animals. Petroleum ether, chloroform (100, 200 mg/kg) and methanol (200 mg/kg) stem extracts didn’t show significant antipyretic activity when compared with the control group of animals (Fig. 2; Table No.2).

Stem bark

In Brewer’s yeast induced pyrexia method, standard group of animals were exhibited antipyretic activity when compared with the control group of animals. Petroleum ether, chloroform and methanol (100, 200 mg/kg) bark extracts didn’t show significant antipyretic activity when compared with the control group of animals (Fig. 3; Table No.3).
Fever may result from infection, one of the sequelae of tissue damage, inflammation, graft rejection, or other disease states. It is produced by certain endogenous substances which include tumour necrosis factor-a (TNFa) and prostaglandins27. Antipyratics have been shown to suppress fever either by inhibiting prostaglandin synthetase, resulting in the blockade of the synthesis of prostaglandin in the brain or suppressing the rise of interleukin-1 production subsequent to interferon production28. The antipyretic potentials of plant may also be attributed to the inhibition of the expression of cyclooxygenase type II (COX-2) which in turn reduces or inhibits the synthesis of PGE 2 in the animals24,29. Antipyretic activity was evaluated for leaves, stem and stem bark in the present investigation by Brewer’s yeast induced pyrexia in experimental rats. In leaves, petroleum ether, chloroform and methanol extracts (100, 200 mg/kg each) exhibited antipyretic activity when compared with the control group of animals. While in stem, methanol (100 mg/kg) extract was exhibited significant (P < 0.05), while petroleum ether, chloroform (100, 200 mg/kg each) and methanol (200 mg/kg) extracts didn’t show significant. Similarly, petroleum ether, chloroform and methanol (100, 200 mg/kg) bark extracts exhibited antipyretic activity.

Alkaloids have been implicated to have the ability to block and inhibit the synthesis of prostaglandin E230 which eventually reduce elevated body temperature in animals. Similarly, flavonoids have been shown to exert antipyretic effect by suppressing TNF-a31. Therefore, the antipyretic activity of the solvent extracts of Pterospermum canescens Roxb., may be associated with pnenolic compounds, flavonoids and or the alkaloid components27 of the plant extracts. It may be inferred that the antipyretic activity of the solvent extracts are the same and may be due to the presence of bioactive agents like flavonoids or alkaloids rather than the quantity of thebioactive priciple(s) present in the plant. The plant extracts may be involved in the inhibition of some of these substances inducing fever.

These experimental results have established a pharmacological evidence for the folklore claim of the drugs to be used as an antipyretic agent. However, further investigation is required to isolate the active constituents responsible for these activities and to elucidate the exact mechanisms of action. The antipyretic activities of Pterospermum canescens Roxb., supports it use in the management of fever by traditional medicine practitioners.

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References


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