Analgesic, Antiinflammatory Activity of Tinospora Cordifolia (Guduchi) and Valeriana Wallichi (Tagara) In Albino Rats

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Abstract

Objectives: The present study was carried out 1) To evaluate analgesic and anti-inflammatory activity of T.Cordifolia and V.Wallichi in albino rats. 2) To compare the analgesic and anti-inflammatory activity of T.cordifolia and V.wallachi with standard drugs pentazocine & diclofenac sodium respectively.

Methods: Analgesic activity of marketed preparation of T.cordifolia, V.wallachi (Guduchi, Tagara from Himalaya Herbal products) was evaluated by tail flick method using analgesimeter. Antiinflammatory activity was studied by rat paw edema method, using plethysmograph. Male wistar albino rats weighing 200 -250g are divided into 8 groups with each group containing six rats (n=6). Test drugs (100mg/kg of T.cordifolia + 120mg/kg of V.wallachi) were administered orally & standard drugs pentazocine 2mg/kg & diclofenac sodium 20mg/kg were given intraperitoneally. One way ANOVA was used for statistical calculation at 5% significance level.

Results: Both test drugs have shown significant analgesic activity (P<0.05), 100mg of T.cordifolia demonstrated significantly higher effect than V.wallachi. And both test drugs has significant anti-inflammatory activity, which was identical.

Conclusion: Tinospora cordifolia & Valeriana wallachi showed significant analgesic and anti-inflammatory activity.

Keywords: Tinospora cordifolia, Valeriana wallachi, Pentazocine, Diclofenac sodium

I. Introduction

The international association of study of pain definition states “pain is an unpleasant sensory and emotional experience associated with actual or potential tissue damage or described in terms of such damage.”[1] Pain is a major symptom in many medical conditions and significantly interferes with a person’s quality of life and general functioning. Analgesics are drugs that selectively relieve pain by acting on the central nervous system or peripheral pain mechanisms without altering the consciousness. Non-steroidal anti-inflammatory drugs and opioid analgesics are the most commonly used drugs for symptomatic relief of pain.

Inflammation is the complex biological response of vascular tissues to harmful stimuli, such as pathogens, damaged cells, or irritants. It is a complex reaction in tissues that consists mainly of response of blood vessels and leukocytes. It is a series of host responses directed as a protective attempt by the organism to remove the injurious stimuli as well as initiate the healing process for the tissue. [2]

Most of the drugs used at present for analgesic effect are synthetic in nature and prolonged use of these drugs causes severe side-effects. [3] In this context, there arises new scope for evaluation of herbs for treatment of pain. Plants still remain a large untapped source of structurally novel compounds that might serve as a lead for the development of novel drugs.

Tinospora cordifolia (common name: Guduchi), is an herbaceous plant of the family Menispermaceae indigenous to the tropical areas of India, Nepal and Sri Lanka. It is known for its immunomodulatory, antioxidant and antibacterial properties. [4][5][6] Tinospora cordifolia is reported to possess antispasmodic, anti-inflammatory, antiallergic, antipyretic, antileptepic and anti-diabetic properties.

Valeriana wallichii (common name: Grandhi Tagaramu), is a pungent perennial herb of the family Valerianaceae that is widely distributed in various temperate regions of globe, including Asia, Europe, and North America, for example. It is one of the most widely used herbal medicines in the world. [7] Valerian root contains bicyclic monoterpenes (valepotriates – notably valtrate and dihydrovaltrate), volatile oils (valeranone, valerenal, and valerenic acids), sesquiterpenes, and alkaloids. [8][9]
II. Aim and Objectives:

1) To evaluate analgesic and anti-inflammatory activity of Tinospora cordifolia and Valariana wallachi in albino rats. 2) To compare the analgesic and anti-inflammatory activity of Tinospora cordifolia and Valareiana wallachi with standard drugs pentazocine & diclofenac sodium respectively.

III. Materials and Methods:

The animals used for the study were male Albino rats (150-250 g). Animals are housed at Central Animal House of Dr. Pinnamenani Siddhartha Medical College & Research foundation which is maintained under standard conditions. The herbal preparation used for this study is taken from Himalaya Herbal Healthcare, in which extracts of these herbal capsules are available.

3.1 Equipments:

Digital analgesiometer (INCO), Plethysmograph (MKM), Mercury, Insulin syringes, Tuberculin syringes, Infant feeding tube, Measuring jar, Glass beakers, Animal weighing balance, Animal cages and water bottles, Cotton, Spirit, Stopwatch, Glass rod, Disposable needles

3.2 Drugs:

The standard drug: Pentazocine 2mg/kg & diclofenac sodium 20mg/kg were dissolved in distilled water and given intraperitonially. Test drug: T.cordifolia (TD1) & V.wallachi (TD2) powder were dissolved in water and administered in an oral dose of 100 and 120 mg/kg respectively.

3.3 The Tail Flick Method:

The tail flick procedure was originally described by D’amor&smith[11] (1941) for testing analgesics in both rats and mice. Male albino rats are selected for the experiments. Animals were weighed with the help of weighing machine. The animals weighing 250gms on average are selected for the experiment. The animals were divided into 3 groups. Each group contains 6 animals. For identification each group was marked with different colours. A portion of the tail is darkened, using ink, at approximately 3 cm from the tip of the tail. Control group of animals were marked with black ink, standard group of animals were marked with blue ink and Test group of animals were marked with red ink. Prior to the experiment all animals normal reaction time for heat on analgesiometer was tested for at least 5 times and reaction time was tabulated. The timer in the analgesiometer will automatically record the tail flick latency. The instrument was operated at 2.5 amps current throughout the experiment.

The rat is inserted in the metallic rat holder and the tail of the rat is taken out from the slit provided in the rat holder. The tail of the rat is positioned in the groove provided. The mains plug is inserted into the mains socket for powering the analgesiometer. The set current knob is rotated anti-clock wise fully. The instrument is switched ON with the help of switch marked with mains on the front panel. Mains ON indicator starts glowing. Current meter will start indicating current in the meter. Now the desired level is set by observing the color of the wire connected between the two terminals below the tail of the rat. It shall be near to red hot. The flicking of the tail of the rat is observed and the time taken for flicking of the tail after heat is applied is noted. A cut off period of 20 seconds is observed to prevent damage to tail. Any animal failing to withdraw its tail within cut off period is rejected from the study. At least 3-5 reading were taken for each rat at a gap of 5 min to the normal behavior of the animal.

3.4 Carrageenan induced paw edema model:

To study the acute and sub acute phases of inflammation in rats. Carrageenan is a widely used irritant or inflammagon or a phlogistic agent. Chemically, it is a sulphated polysaccharide obtained from sea weed (rhodophyceae). The experimental tissue injury caused by this irritant initiates a cascade of inflammatory events leading to formation of exudates. The inflammation induced by it is biphasic in nature. The first phase is attributed to the release of histamine, 5-hydroxy tryptamine (serotonin) and kinin while the second phase is related to the release of prostaglandins. The well recognized method of winter et al, 1962 is followed.[12] A 1% w/v suspension of carrageenan is prepared freshly in normal saline and injected into sub planter region of left hind paw (usually 0.1ml in rats). In control group animals only normal saline is injected. Test drugs T.cordifolia & V.wallachi powder were dissolved in water and usually administered orally, according to body weight, half an hour before the carrageenan challenge. A mark is made at the ankle joint (tibio-tarsal joint) of each rat. Paw edema volume up to the ankle joint is measured in drug treated and untreated groups at 0 and at 3hours following carrageenan challenge by using mercury Plethysmograph filled with mercury.[13] Percentage (%) of reduction in edema is calculated.

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3.5 Grouping:
There are eight groups each group consist of n=6 rats total n=48 rodents.

- **Group 1** - Administered with 0.5 ml of normal saline.
- **Group 2** - Administered with 2 mg/kg body weight (BW) of pentazocine.
- **Group 3** - Administered with the extract of T. cordifolia (TD1) 100 mg/kg BW.
- **Group 4** - Administered with the extract of V. wallichii (TD2) 120 mg/kg BW are tested for analgesic effect.
- **Group 5** - Administered with 0.5 ml of normal saline.
- **Group 6** - Administered with 20 mg/kg body weight (BW) of diclofenac sodium.
- **Group 7** - Administered with the extract of T. cordifolia (TD1) 100 mg/kg BW.
- **Group 8** - Administered with the extract of V. wallichii (TD2) 120 mg/kg BW are tested for anti-inflammatory effect.

3.6 Statistical analysis:
The results are expressed by taking tail flick latency time and percentage of inhibition in rat paw edema. Statistical analysis is performed using one way ANOVA at 5% significance level.

### IV. Figures And Tables:

#### Table 1: comparison of mean reaction time of Normal Saline, Pentazocine, and Tinospora & Valeriana at 0 min, 30 min, 60 min & 90 min.

<table>
<thead>
<tr>
<th>Group</th>
<th>Dose</th>
<th>Mean reaction Time in minutes</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
</tr>
<tr>
<td>I</td>
<td>Normal saline - 5ml/kg</td>
<td>7.63±0.46</td>
</tr>
<tr>
<td>II</td>
<td>Pentazocine - 2mg/kg</td>
<td>7.47±0.21</td>
</tr>
<tr>
<td>III</td>
<td>T.cordifolia - 100mg/kg</td>
<td>7.17±0.75</td>
</tr>
<tr>
<td>IV</td>
<td>V.wallichii - 120mg/kg</td>
<td>7.23±0.84</td>
</tr>
</tbody>
</table>

The mean reaction time in control group at 0 min was 7.63±0.46, at 30 mins 7.52±0.56, at 60 mins 7.47±0.21, at 90 mins 7.50±0.55. For pentazocine group the mean reaction time at 0 min was 7.47±0.21, at 30 mins 11.5±1.05, at 60 min 13±1.27, at 90 mins 14.83±0.98 respectively. The mean reaction time of Tinospora cordifolia in dose of 100mg/kg at 0 min was 7.17±0.75, at 30 mins 8.67±0.82, at 60 mins 10.83±0.75, at 90 mins 13.17±0.75. The mean reaction time of Valeriana wallichii in dose of 120mg/kg at 0 min was 7.23±0.84, at 30 mins 8.14±0.71, at 60 mins 9.58±0.58, at 90 mins 12.84±0.64 respectively. The mean reaction time increased significantly with T.cordifolia and V.wallichii at doses of 100 & 120mg/kg when compared to control. Both the test drugs show statistically significant analgesic activity, but when compared with each other T.cordifolia showed more analgesic effect.

#### Table 2: comparison of mean increase in paw volume of Normal Saline, Diclofenac sodium, Tinospora cordifolia and Valeriana wallichii

<table>
<thead>
<tr>
<th>Groups</th>
<th>Dose</th>
<th>Mean Increase in Paw Volume in ml at hours</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0hr</td>
</tr>
<tr>
<td>I</td>
<td>Normal saline - 5ml/kg</td>
<td>4.11±0.04</td>
</tr>
<tr>
<td>II</td>
<td>Diclofenac sodium - 20mg/kg</td>
<td>4.12±0.03</td>
</tr>
<tr>
<td>III</td>
<td>T.cordifolia - 100mg/kg</td>
<td>4.13±0.05</td>
</tr>
<tr>
<td>IV</td>
<td>V.wallichii - 120mg/kg</td>
<td>4.12±0.04</td>
</tr>
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</table>

In rat paw volume method, for normal saline the volume of paw edema at 0 hr was 4.11ml and increased to 4.37ml after 3 hrs. In case of diclofenac sodium (standard drug), the volume of paw edema at 0 hr was 4.12ml and increased to 4.23ml after 3 hrs. In case of T.cordifolia and V.wallichii, the volume of paw edema at 0 hr was 4.13ml and 4.12 and increased to 4.3ml and 4.28 ml after 3 hrs respectively. So, here both the test drugs showed significant anti-inflammatory activity when compared to control. The present study suggests the possible potential role of Tinospora cordifolia and Valeriana wallichii as an analgesic and anti-inflammatory drug, however further research is required to establish its use.
Analgesic, Antiinflammatory Activity of Tinospora Cordifolia (Guduchi) and Valeriana Wallichii.

EFFECT OF DRUGS ON RAT PAW VOLUME

VI. Discussion

In spite of the availability of various anti-inflammatory agents, we were interested in evaluating the herbal drugs which can be widely used for different therapeutic purposes. The herbal formulations are relatively free from adverse effects and chances of drug dependence with them are very less. The study comprises comparison of analgesic & anti-inflammatory effect of Tinospora cordifolia (guduchi) and Valariana wallichii (tagara) in rats by using tail flick method and Rat paw edema method respectively. Our study is supported by Siddalingapa et.al.,(2011) they evaluated analgesic and anti-inflammatory activity of aqueous extract of Tinospora cordifolia (AECT) in rodents, by using two different analgesic and anti-inflammatory methods. Their observations also indicated that the extract have both analgesic and anti-inflammatory effects. The finding of the study conducted by Sangeetha PikhwalSah et, al.,(2010) and Supriya Priyambada et,al, (2015) were in conformity with our study i.e., Valariana wallichii extract has potent analgesic and anti-inflammatory activity in tail flick and carrageenan induced inflammation in albino rats. The current study demonstrates significant (p<0.001) analgesic activity evidenced by increase in the reaction time by Tail flick method and anti-inflammatory activity by reduction in carrageenan induced paw edema method. The percentage of rat paw inhibition of standard (SD) and test drugs (TD1, TD2) as compared to control was 38%, 31% and 33% respectively.

VI. Summary And Conclusion:

Tinospora cordifolia and Valariana wallichii was found to be having analgesic and anti-inflammatory property at 100mg/kg and 120mg/kg dose. The results were assessed and the mean, SD and percentage inhibition was noted. The statistical analysis was done by one way ANOVA test. The current study demonstrates significant analgesic and anti-inflammatory effect of T.cordifolia and V.wallichia, but they are
statistically less significant when compared to standard drug pentazocine and diclofenac. T. cordifolia demonstrated significantly higher analgesic effect than V. wallachi. And both test drugs has significant anti-inflammatory activity, which was identical.

**Bibliography**


