EVALUATION OF PHYTOCHEMICAL INVESTIGATION AND ANTICONVULSANT ACTIVITY OF AQUEOUS STEM EXTRACT OF ECLIPTA ALBA

Mahanthesh M.C*, Swati MP, Popat SK, Vikram HP, Jadhav SY, J.I.Disouza

Department of Pharmacognosy, Tatyasaheb Kore College of Pharmacy, Warananagar 416113

ABSTRACT

The present study expose that the anticonvulsant activity of Eclipta Alba (Asteraceae) stem extracts. The anti-convulsant activity of plant extracts was evaluated by using the Maximal Electroshock (MES) induced convulsion technique. The convulsions in experimental animals were induced with Maximal electroshock of 150 mA current for 0.2 sec. Then administration of normal saline, standard drug and stem extracts at respective doses. The stem extract exhibited a significant reduction in various phases of epileptic seizure. The aqueous extracts treated groups showed significant decrease in duration of flexion; clonus and stupor phase of MES induced convulsions. The Eclipta alba possessed significant anticonvulsant activity against MES and PTZ induced convulsions.

KEY WORDS: MES, Phenytoin, Diazepam, Eclipta alba, Asteraceae.

INTRODUCTION

Epilepsy is the common chronic central nervous system disorder characterized by transient signs and symptoms of abnormal, excessive or synchronous neuronal activity in the brain. (Fisher etal 2005) It is a major neurological disorder and up to 1-2 % of the world population develops epilepsy in their lifetime was reported (Reinaldo etal 2011).The current therapy of epilepsy with modern antiepileptic drugs is associated with side effects, dose-related and chronic toxicity, as well as teratogenicity effects, and approximately 30% of the patients continue to have seizures with current antiepileptic drugs therapy (CascinoGD 1994).Traditional systems of medicine are popular in developing countries and up to 80% of the population relies on traditional medicines or common remedies for their primary health
Eclipta alba is commonly known as False Daisy or Bhringaraj. It is a creeping and moisture loving herb commonly found on roadsides and waste lands throughout India. The plant is known to have some important pharmacological activities such as hepatoprotective, antimicrobial, antinociceptive, analgesic, antiinflammatory, antiviral, immunomodulatory and nootropic activity (Thorat et al 2010). Several plants used for the treatment of epilepsy in diverse systems of traditional medicine. The present study covers Phytochemical investigation and evaluation of anticonvulsant activity using maximal electrode shok (MES) and pentaleentetrazole (PTZ) indused convulsions.

MATERIALS AND METHODS

![Eclipta alba](image)

Figure No. 01 *Eclipta alba*

**Synonyms** : EcliptaErecta, Eclipta alba, Verbesina alba, Verbecina Prostrata, wedeliacalendulacea

**Binomial name** : Eclipta prostate

**Family** : Asteraceae

**Kingdom** : Plantae

**Order** : Asterals

**Genus** : Eclipta (Thorat et al 2010)

**Collection and authentication of plant material**

The fresh stem of Eclipta alba were collected in the month of September from surrounding area of Warananagar, Kolhapur district, Maharashtra. India. The plant was positively identified and confirmed by botanist Dr. S. Y. Jadhav. Head of the department of Botany. Yashwantrao Chavan Mahavidylaya. Warananagar. The plant material was thoroughly
washed with tap water and kept for drying in shade at room temperature and thoroughly air
dried plant material was grinded to powder, and it was passed through the sieve no. 40.
Subjected to maceration process. The fine particle was collected. Then it was poured into the
conical flask and added a sufficient amount of water and chloroform. Chloroform was added
because to avoid the microbial growth into aqueous extract. The flask is allowed to stand for
7 days. It to be shooked well every day.

**Phytochemical Screening** (Kokte CK 2006)

**Table 01: Preliminary Phytochemical screening of *Eclipta alba* stem aqueous extract**

<table>
<thead>
<tr>
<th>Sr. no.</th>
<th>Test Name</th>
<th>Observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Test for alkaloids</td>
<td>(+ve)</td>
</tr>
<tr>
<td>2.</td>
<td>Test for glycoside</td>
<td>(+ve)</td>
</tr>
<tr>
<td>3.</td>
<td>Test for triterpenoids</td>
<td>(+ve)</td>
</tr>
<tr>
<td>4.</td>
<td>Test for tannin</td>
<td>(+ve)</td>
</tr>
<tr>
<td>5.</td>
<td>Test for saponin</td>
<td>(+ve)</td>
</tr>
<tr>
<td>6.</td>
<td>Test for phenols</td>
<td>(+ve)</td>
</tr>
</tbody>
</table>
Table 2: The Effect of MES Induced convulsions of Aqueous Stem of *Eclipta Alba*

<table>
<thead>
<tr>
<th>Drug</th>
<th>Dose mg / kg b.w</th>
<th>Route of Administration</th>
<th>Time (Sec) in various phases of convulsions (Mean ± SEM)</th>
<th>Recovery/Death</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Salaine)</td>
<td></td>
<td>Oral</td>
<td>Flexion: 3.83±0.6, Extensor: 11.16±1.60, Clonus: 3.50±0.88, Stupor: 110.66±6.92</td>
<td>Recovery</td>
</tr>
<tr>
<td>Standard Phenytoin</td>
<td>25</td>
<td>Intra peritoneal (i.p)</td>
<td>4.5±0.76, 00±00, 00±00, 00±00</td>
<td>Recovery</td>
</tr>
<tr>
<td>Aqueous</td>
<td>200</td>
<td>Oral</td>
<td>3.50±088, 00±00, 4.50±0.99, 40.66±9.40</td>
<td>Recovery</td>
</tr>
</tbody>
</table>

Mean ± SD, p<0.0001, Considered extremely significant on comparing with the normal group. One Way ANOVA

Table 3: The Effect of PTZ Induced convulsions of Aqueous Stem of *Eclipta Alba*

<table>
<thead>
<tr>
<th>Drug</th>
<th>Dose mg / kg b.w</th>
<th>Route of Administration</th>
<th>Onset of convulsions (Sec. ± SEM)</th>
<th>Mortality %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control PTZ</td>
<td>80</td>
<td>Intra peritoneal (i.p)</td>
<td>78.66±6.29, 0/6, 6/6</td>
<td>100.0%</td>
</tr>
<tr>
<td>Standard Diazepam +PTZ</td>
<td>4+80</td>
<td>Intra peritoneal (i.p)</td>
<td>00±00, 6/6, 0/6</td>
<td>0%</td>
</tr>
<tr>
<td>Aqueous</td>
<td>200</td>
<td>Oral</td>
<td>177.50±8.24, 6/6, 3/6</td>
<td>0%</td>
</tr>
</tbody>
</table>
Thin layer chromatography of *Eclipta alba*

![TLC image](image)

**Figure No. 02 TLC of Eclipta alba (Ethyl acetate: Methanol (16:4))**

The thin layer chromatography of aqueous extract was carried out using

**Mobile phase**: Ethyl acetate: Methanol (16:4).

**Stationary phase**: Silica gel.

**Developing agent**: iodine vapors (Kasture *et al* 2012)

**Macroscopic and Microscopic Examination**

Transverse section of stem shows epidermis, composed of single layered epidermis, externally covered with cuticle, a few epidermal cells elongate to form characteristic non-glandular trichomes, the cork where formed, poorly developed consisting of rectangular cells; secondary cortex composed of large, rounded or irregular shaped parenchymatous cells having wide air spaces; endodermis single layered consists of tangentially elongated cells; Pericyclic fibres distinct, arranged in tangential strands; vascular bundles in a ring, collateral, end arch, of varying sizes traversed by medullary rays; phloem a narrow strip composed of sieve elements and phloem parenchyma; xylem consists of large number of vessels, xylem fibres and xylem parenchyma; xylem vessels appear evenly distributed throughout the xylem; in macerated preparation vessels barrel-shaped, some elongated with simple perforations, pitted with spiral thickening; xylem fibers with wide lumen, pointed tips and pitted walls, a few often bifurcate and a few other large, peg-like outgrowth; xylem parenchyma rectangular with pitted thickening; xylem rays triseriate to pentaseriate, normally biseriate and uniseriate, 8-15 cells in height and 3-5 cells in width; center occupied by a wide pith consisting of isodiametric cells.(Khandelwal 2005)
Evaluation of Anticonvulsant Activity (Kulkarni SK)

Male wistar rats weighing 200-300 gm of either sex were procured from animal house of the Krishna Institute of Medical Science, Karad (approved by the animal Ethical committee). All the animals are kept in standard polypropylene cages and maintained under standard conditions like temperature (24 ± 10 C), relative humidity (45-55 %) and 12:12 light: dark
cycle. The animals were fed with standard rat pallet and water. The animals were allowed to acclimatize to laboratory conditions 48 hrs before the start of the experiment. Rats were used for determination of acute toxicity by fixed dose method (Reed and Meunch). Groups of 6 rats (200-300 gm.) were used in all sets of experiments. Group –I received tween 80, (5ml/kg), served as solvent control. Group II received diazepam, served as control. Group III and IV received methanol extract of Eclipta Alba. All the experiments were conducted after obtaining permission from the Institutional Animal Ethics Committee (IAEC) Experimental convulsion, in albino rats was induced by intra peritoneal administration of pentylenetetrazole (PTZ) at 80mg/kg, in sterile distilled water after half an h administration of test drug. Then onset of clonic convulsions were estimated and compared with the solvent control group. The standard drug diazepam 4mg/kg, dissolved in sterile distilled water was used as control.

RESULTS AND DISCUSSION
The results of the present study indicate that Aqueous Stem extract of Eclipta alba (ASEAA) possesses anticonvulsant activity in rats. GABA is the major inhibitory neurotransmitter in the brain while glutamic acid is an excitatory neurotransmitter in the brain. The inhibition of GABA neurotransmitter and the enhancement of the action of glutamic acid have been shown to be the underlying factors in epilepsy. Our study shows that the aqueous extract of the stem of Eclipta alba protected some of the animals against seizures induced by maximal electroshock (MES), pentylenetetrazole (PTZ) and also delayed the latency of the seizuresTable 2and 3. As shown in table 2, aqueous extract of stem of Eclipta alba at doses of 200 mg/kg and Phenytoin (25 mg/kg) have shown significant reduction (p<0.001) in duration of convulsions. The aqueous extract has good anticonvulsant activity. Antiepileptic drugs that block MES-induced tonic extension are known to act by blocking seizure spread. Moreover, drugs that inhibit voltage-dependent Na+ channels, such as phenytoin can prevent MES-induced tonic extension.

Pentylenetetrazole(PTZ) induced seizures in all the rats used. Pentylenetetrazole may elicit seizures by inhibiting gabaergic mechanisms. Standard antiepileptic drugs, diazepam and phenobarbitone, are believed to produce their effects by enhancing GABA mediated inhibition in the brain. It is, therefore, possible that the anticonvulsant effects shown in this study by the drugs against seizures produced by PTZ might be due to the activation of GABA neurotransmission. Since the extract similarly antagonized seizures elicited by
pentylenetetrazole in rats, it is probable, therefore, that it may also be exerting its anticonvulsant effects by affecting gabaergic mechanisms (Karunakar et al 2009). The phytochemical screening of the extract revealed the presence of small quantities of alkaloids, flavonoids, saponins and large amounts of cardiac glycosides, triterpenoids, phenolic compounds and tannins (Table.1). Based on the present state of knowledge of the chemical constituents of the extract, it is not possible to attribute with certainty its anticonvulsant effect to one or several active principles among those detected in the screening, are reported to possess anticonvulsant activity in experimental seizure models such as MES and PTZ.

CONCLUSION
It can be concluded from the study that, the anticonvulsant effects of the Aqueous Stem extract of *Eclipta alba* (ASEAA) may be via non-specific mechanisms. However, extensive studies are needed to evaluate the precise mechanism(s), active principles, and the safety profile of the plant as a medicinal remedy for convulsive disorders.

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REFERENCE


