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EVALUATION OF ANTIEPILEPTIC ACTIVITY OF AQUEOUS EXTRACT OF LEAVES OF GOSSYPIUM HERBACEUM IN MICE

Sumalatha G[1] and Sreedevi A[2]*

[1]*Vaagdevi College of Pharmacy, Hanamkonda, Andhra Pradesh*

[2]*Institute of Pharmaceutical Technology, Tirupati, Andhra Pradesh*

Corresponding author E mail: sridevitirupati@rediffmail.com

ABSTRACT

Aim:*The aim of the present study was to investigate antiepileptic activity of aqueous extract of Gossypium herbaceum (AEGH) in mice.* ***Method:****The antiepileptic activity of AEGH at 10, 30, and 100 mg/Kg, p.o. was evaluated by the convulsions induced in mice by maximum electroshock (MES), Pentylenetetrazole (PTZ) and Isoniazid (INH). Statistical analysis was carried out by one-way analysis of variance (ANOVA) followed by Dunnett's t test.* ***Results and Discussion:*** *In MES method, AEGH (10, 30, and 100 mg/Kg) inhibited convulsions significantly potent than Diazepam. In PTZ method, AEGH inhibited convulsions potent than Phenobarbitone sodium (PS). In INH method, AEGH delayed the onset of convulsions less potent than Diazepam.* ***Conclusion:*** *In Present investigation, AEGH showed significant dose dependent antiepileptic effect potent than Diazepam and PS.*

KEYWORDS

Epilepsy, Pentylenetetrazole, Isoniazid.

INTRODUCTION

Epilepsy is most common neurological disorders affecting people across all nationalities[1]. The word epilepsy in derived from the Greek verb **epilamvanein** (to be seized", "to be taken hold off", or "to be attacked" indicating that the person having a seizure is 'possessed' or at least out of control[2]. Epilepsy refers to chronic conditions characterized by recurrent seizures[1]. Epilepsy is one of the most common neurological disorders characterized by sudden, transient alterations of brain function usually with motor, sensory autonomic or psychic symptoms often accompanied by loss of, or altered consciousness. Several biochemical hypotheses suggest the involvement of decreased activity of inhibitory GABA ergic system or increased activity of excitatory amino-acids (glutamate and aspartate system) in epilepsy[2].

Epilepsy is treated mainly with drugs; though brain surgery may be used for severe cases. The antiepileptic drugs (AED's) like valproate, phenytoin and carbamazepine are associated with osteoporosis and other disorders of bone and mineral metabolism[3]. In addition use of antiepileptic drugs during pregnancy increases the risk for specific congenital malformations such as neural tube defects, cleft lip and palate and cardiovascular malformations[4]. Even the current antiepileptic drugs such as Oxcarbazepine, gabapentin, tiagabine, topiramate, levotiracetam, lamotrigin, felbamate, and fosphenytoin have the drawbacks like limited spectrum or drug

interactions with oral contraceptives. It is felbamate and lamotrigine that have potential of significant side effects[5]. The currently used antiepileptic drugs fail to provide satisfactory seizure control and toxicities associated with these drugs can further compromise quality of life while drug-drug interactions may complicate clinical management.

Keeping these complications in mind, various herbal medicines have been tried in the past for their potent anticonvulsant properties. *Gossypium herbaceum* is a shrub belonging to the family Malvaceae. Traditionally, flowers were used as anti inflammatory and roots were used as diuretic agents. Root bark of *Gossypium herbaceum were* given with water for miscarriage[6]. Cotton seeds were used to treat epilepsy[7]. The aim of the present study is to evaluate the potential of aqueous extract of leaves of *Gossypium herbaceum (*AEGH) to protect the mice from convulsions.

MATERIALS AND METHODS

Plant Material

Leaves of *Gossypium herbaceum* were collected from Nellutla, Andhra Pradesh, India. It was identified and authenticated by Prof. V. S. Raju, department of Botany, Kakatiya University.

Preparation of the extract

The fresh leaves of *Gossypium herbaceum* were collected and washed under running tap water. They were shade dried at room temperature. Then dried leaves were made in to coarse powder. The powder was passed through a 60 No mesh sieve. Then aqueous extract was prepared by by cold maceration[8].

Qualitative analysis

The extract was subjected to phytochemical screening by using different qualitative tests[9].

Acute toxicity study

Acute toxicity study will be performed for the extracts to ascertain safe dose by acute oral toxic class method of Organization of Economic Co-operation and Development, as per 420 guidelines (OECD) [10].

Antiepileptic activity

Maximum electroshock (MES) in mice

Five groups of six male Swiss albino mice (25 – 30) were used. The test was started one hour after oral treatment with the test compound (AEGH 10, 30, and 100 mg/Kg, p.o.) or the vehicle or the standard (Diazepam 3 mg/Kg, p.o.). An apparatus with corneal electrodes was used to deliver the stimuli. The intensity of the stimulus is dependent on the apparatus, eg: 30mA, 50Hz for 0.2 sec has been used. The onset and the duration of tonic himb extension was recorded and percentage of inhibition of seizures relative to controls was calculated[11].

Pentylenetetrazole (PTZ) induced convulsions in mice

Control group received vehicle, test group received AEGH (10, 30, and 100 mg/Kg, p.o.) and standard group received Phenobarbitone sodium, (40 mg/Kg, i.p.). Convulsions were induced by administering PTZ (75 mg/Kg, i.p.), 1hr after AEGH and 15 min after PS and diazepam administration. The onset and the duration of convulsions were recorded and percentage inhibition was calculated[12].

Isoniazid (INH) induced convulsions in mice

Control group received vehicle, test group received AEGH (10, 30 and 100 mg/Kg, p.o.) and standard group received Diazepam, (4 mg/Kg, i.p.). Convulsions were induced by administering INH (300 mg/Kg, s.c.), 1hr after drug administration. The onset time of convulsions was recorded[13].

STATISTICAL ANALYSIS:

The data were analyzed by using one-way analysis of variance (ANOVA), followed by Dunnett's test. P <0.05 was considered as statistically significant.

RESULTS AND DISCUSSION

Qualitative analysis

In preliminary phytochemical screening aqueous extract shows presence of steroids, alkaloids, glycosides, tannins and proteins.

Acute toxicity study

Toxicity was found at 1 gm/Kg, p.o. So, 10, 30 and 100 mg/Kg were the doses selected for the study.

Antiepileptic activity

Maximum electroshock (MES) in mice

AEGH exhibited significant dose dependent antiepileptic activity. AEGH at both 100 mg/Kg exhibited antiepileptic activity potent than Diazepam. (**Table 1**)

Pentylenetetrazole (PTZ) induced convulsions in mice

AEGH exhibited significant dose dependent antiepileptic activity. AEGH at both 30 and 100 mg/Kg exhibited antiepileptic activity potent than PS. (**Table 2**)

Isoniazid (INH) induced convulsions in mice

AEGH at all three doses exhibited significant dose dependent delay in onset of convulsions less potent than Diazepam. (**Table 3**)

Table 1: Effect of AEGH on maximal electroshock induced convulsions in mice

Treatment	Dose	Onset time (s)	Duration of tonic hind limb extension (s)	Percentage of Inhibition (%)
Vehicle	1 ml	1.51±0.09	93.55±0.17	-
AEGH	10 mg/Kg	1.89±0.07**	43.66±0.06**	53.33**
AEGH	30 mg/Kg	3.32±0.13**	39.84±0.16**	57.42**
AEGH	100 mg/Kg	5.06±0.11 **	35.54±0.12**	62.01**
Diazepam	3 mg/Kg	3.55±0.19**	39.56±0.09**	57.71**

AEGH: Aqueous extract of leaves of *Gossypium herbaceum* ; Values are mean ± SD (n = 6). Statistical significance was determined by ANOVA, followed by Dunnett's t test (n=6); $^{**}P < 0.01$.

Table 2: Effect of AEGH on PTZ-induced convulsions in mice

Treatment	Dose	Onset time (min)	Duration of convulsions (min)	Percentage of Inhibition (%)
Vehicle	1 ml	7.51±0.05	19.23±0.07	-
AEGH	10 mg/Kg	13.35±0.05**	12.47±0.04**	35.14**
AEGH	30 mg/Kg	18.29±0.05**	8.38±0.05**	56.41**
AEGH	100 mg/Kg	23.39±0.04**	3.34±0.03**	82.65**
Phenobarbitone sodium	300 mg/Kg	4.49±0.04**	9.14±0.04**	52.46**

AEGH: Aqueous extract of leaves of *Gossypium herbaceum* ; Values are mean ± SD (n = 6). Statistical significance was determined by ANOVA, followed by Dunnett's t test (n=6); $^{**}P < 0.01$.

Table 3: Effect of AEGH on INH-induced convulsions in mice

Treatment	Dose	Onset of convulsions (min)
Vehicle	1 ml	24.14±0.04
AEGH	10 mg/Kg	29.55±0.03**
AEGH	30 mg/Kg	35.12±0.08**
AEGH	100 mg/Kg	40.55±0.03**
Diazepam	4 mg/Kg	63.27±0.13**

AEGH: Aqueous extract of leaves of *Gossypium herbaceum*; Values are mean ± SD (n = 6). Statistical significance was determined by ANOVA, followed by Dunnett's t test (n=6); $^{**}P < 0.01$.

CONCLUSION

In the present investigation, AEGH showed significant dose dependent antiepileptic effect potent than Diazepam and PS which may be due to enhanced GABA ergic neurotransmission.

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***Corresponding Author:**
Sreedevi A*
Institute of Pharmaceutical Technology,
Tirupati, Andhra Pradesh.
E-mail: sridevitirupati@rediffmail.com