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(RESEARCH ARTICLE)

# Phytochemical and neuropharmacological activity of leaves extract of *Tridax procumbens* linn

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## Abstract

The neuropharmacological activities of the hydroalcoholic extract of Tridax procumbens were screened in mice. Preliminary phytochemical evaluation of extract was also carried out. The extract (200 and 400mg./kg orally.) possess a significant ($p<0.05$) depression in general behavioral test. The HAETP showed a significant reduction in spontaneous motor activity, rota rod test showed reduction on grip strength was found significant and open field method significantly reduction in number of squares entered with both forelimbs with compare with normal control.

**Keywords:** Tridax procumbens Linn; Diazepam; Rota-rod apparatus; Actophotometer test; Open field test

## 1. Introduction

According to The World Health Organization, depression is among the top ten causes of morbidity, and mortality world over [1], and it is envisaged to be the leading cause of morbidity and mortality by the year 2030 [2]. Patients with depression have symptoms that reflect a functional deficit in brain monoamine neurotransmitters, specifically norepinephrine, serotonergic and dopaminergic systems [3]. Other neurotransmitters such as GABA acting via GABAB receptor, β adrenoceptors, muscarinic cholinergic receptor, glutaminergic via NMDA receptor [4,5], and nitric oxide signaling pathways have been implicated in depression. Despite the availability of synthetic antidepressant drugs, depression remains a major medical problem [6]. This is because the currently available antidepressant drugs are associated with a numerous side effects which include weight gain, hypopiesia, sexual dysfunction, cardiac toxicity and sleep disorder [7,8,9]. Plants are the primary source of medicinal products, which play a key role in global health. Human beings have been using plants for many years to cure and mitigate diseases (10). WHO recorded that more than 75% of people use herbal medicines to meet their everyday health needs (11, 12). Therefore, herbal therapies should be considered alternative or complementary medicines [13].

The Tridax procumbens Linn is a common weed and is found throughout the year, it is commonly known as '' Coat buttons'' belongs to the family Asteraceae. Previous studies have demonstrated that the leaf extracts of this plant contained antidiabetic and anti hyperlipidimic effects, anti-arthritic, anti-cancer activity, analgesic and anti-inflammatory activity, anti-fungal activity, anthelmintic activity and hepatoprotective properties. Many bioactive compounds have been isolated from leaves, aerial parts and flowers including alkaloids, triterpenoids, flavonoids, glycoprotein, carotenoids, saponins and tannins.[14,15,16]. The current investigation focused on the neuro pharmacological benefits of this plant in the in vivo mouse model of Tridax procumbens.

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## 2. Materials and Methods

### 2.1. Plant material

*2.1.1. Collection and Authentication of plant materials:*

The whole plant were collected from the Village side, Sivagangai district and it was confirmed taxonomically and authenticated by Dr.Stephen., Botanist of American college, Madurai, Tamilnadu.

*2.1.2. Preparation of plant material:*

The leaves were separated and washed after then shade dried for 8-10 days. Then the dried leaves are powdered in coarse powder. This powder was stored in an air tight container and used for successive extraction.

*2.1.3. Extraction procedure:*

About 250 gram of coarse powder of *Tridax procumbens* was packed in a soxhelt apparatus and then extracted with petroleum ether by continuous hot percolation method. Then marc 1 left out and then extracted with benzene then Marc 2 extracted with ethanol and distilled water (70: 30) as Polar solvent for a period of 72 hours, at the end of extraction, the extract was concentrated to dry mass by evaporation. After concentration a dark green viscous residue was obtained. The crude extract was stored in a refrigerator.

*2.1.4. Preliminary Phytochemical Screening*

The hydroalcoholic extract of *Tridax procumbens* was screened for the presence of various phytoconstituents like steroids, alkaloids, glycosides, flavonoids, carbohydrates, proteins and phenolic compounds [17].

### 2.2. Experimental animals:

All animal studies were conducted after approval from the Institution of ethical committee (IAEC.No: 11/2017), Madurai Medical College, Madurai. Albino mice (25-30g) were used for the study were housed in care facility Institute of Pharmacology, Madurai Medical College, Madurai. The animals were stored in polypropylene cage (room temperature 25±1°c with 12 hours light & 12 hours dark cycle relative humidity approximately 60°c) with free standard pellets and tap water mice were acclimatized for 10 days before experimentations.

### 2.3. Experimental Design

Six animals were used in each group for each experiment separately. Drugs / vehicle were administered to the animals 30min prior to study.

- Group I: Negative control, administer saline 1 ml/kg orally.
- Group II: Positive control received standard drug Diazepam (4 mg/kg i.p)
- Group III: Received HAETP 200 mg/kg orally
- Group IV: Received HAETP 400 mg/kg orally.

### 2.4. Neuropharmacology activity

*2.4.1. Rota-rod performance*

Four animals at a time were placed on rod rotating at 20–25 rpm speed. Only the mice that demonstrated their ability to remain on the revolving rod (20–25 rpm) for 5 min after training sessions during pretest screening were selected for studies. The fall off time was recorded in all the groups before and 30 min after drug administration. Decrease in fall off time is suggestive of depression of the central nervous system (CNS)[18].

*2.4.2. Actophotometer test*

The animal locomotor behavior was monitored using actophotometer. Animals were placed in actophotometer individually, and basal activity score was recorded over the period of 5 min. Each animal was treated with respective drug, and activity score was recorded after 30 min. Decreased activity score was taken as index of CNS depression [19-20].

*2.4.3. Open field test*

Open field apparatus was designed as described by Gray and Lalji (1971) with few modifications. Dimensions were 50 × 50 × 40 cm made up of plywood open from top and bottom kept on white table top; surface was divided into 25 equal squares, i.e., 9 central and 16 peripheral. During 5-min session of observation, each animal was placed in the corner of open field apparatus, and behavior of animal as determined by ambulation (number of squares entered with both forelimbs), rearing, preening, and defecation was recorded [21].

### 2.5. Statistical Analysis

The experimental data were expressed as mean ± SEM. The significance of difference among the various treated groups and control group were analyzed by means of one-way ANOVA followed by Dunnett's multiple comparison tests. A $p < 0.05$ was considered statistically significant.

## 3. Results

### 3.1. Phytochemical Screening

Phytochemical screening of hydro alcoholic extract of leaves of *Tridax procumbens* Linn revealed the presence of various phytochemical constituents like alkaloids, flavonoids, tannins, saponons, terpenoids and phytosterols.

### 3.2. Rota rod methods.

**Table 1** Mean fall off time in rota-rod method

| S.No | TREATMENT | DOSE | FALL OF TIME (sec) | |
|---|---|---|---|---|
| | | | BEFORE DRUG | AFTER DRUG |
| 1 | Normal control | 1ml/kg | 223 ±11.9 | 226.7±12.3 |
| 2. | Standard - Diazepam | 4mg/kg | 229.7±8.4 | 86.4±2.4* |
| 3. | HAETP | 200 mg/kg | 225±14.5 | 139.6±8.4* |
| 4. | HAETP | 400mg/kg | 220±10.6 | 106.8±6.4* |

Values are expressed as Mean ±SEM (n=6); *P< 0.05 compared with the control group (Dunnett's Test)

Table No.1. Diazepam (4 mg/kg i.p.) and HAETP (200 & 400mg/kg oral.) treated groups showed significant CNS depressant activity 86.4±2.4, 139.6±84, 106.8±6.4 when compared with control {226.7±12.3}.

### 3.3. Actophotometer Test

**Table 2** Activity score in Actophotometer

| S.No | Treatment | Dose | Locomotor activity IN 5MINS (counts) | |
|---|---|---|---|---|
| | | | Before drug | After drug |
| 1 | Normal control | 1ml/kg | 350±12.5 | 346±12.2 |
| 2. | Standard - Diazepam | 4mg/kg | 356±16.4 | 124.2±6.8* |
| 3. | HAETP | 200 mg/kg | 348±14.8 | 168±4.2* |
| 4. | HAETP | 400mg/kg | 354±10,6 | 146.6±2.6* |

Values are expressed as Mean ±SEM (n=6); *P< 0.05 compared with the control group (Dunnett's Test)

Table No.2. Diazepam (4 mg/kg i.p.) and HAETP (200 & 400mg/kg oral.) treated groups showed significant reduction locomotor activity 124.2±6.8, 168±4.2, 146.6±2.6 when compared with control 346±12.2.

### 3.4. Open field test.

**Table 3** Mean score in open field method

| S.No | Treatment | Dose | No. OF CROSSING IN 5MINS (SQUARES CROSSED WITH ALL FOUR LIMBS) | |
|---|---|---|---|---|
| | | | Before drug | After drug |
| 1 | Normal control | 1ml/kg | 101.7±2.5 | 100.6±5.6 |
| 2. | Standard - Diazepam | 4mg/kg | 98±6.4 | 25.2±3.4* |
| 3. | HAETP | 200 mg/kg | 108±4.8 | 62.2±6.2* |
| 4. | HAETP | 400mg/kg | 105±4,6 | 46.6±4.2* |

Values are expressed as Mean ±SEM (n=6); *P< 0.05 compared with the control group (Dunnett's Test)

Diazepam (4mg/kg i.p.) and HAETP (200&400mg/kg oral) significantly decreased in number of crossing 25.2±3.4, 62.2±6.2, 46.6±4.2 compare with normal control 100.6±5.6.

## 4. Discussion

Anxiety and hypnosedation are principally mediated in the CNS by the $GABA_A$ receptor complex, which is also involved in other physiological functions related to behavior and in various psychological and neurological disorders such as epilepsy, anxiety, depression, Parkinson syndrome, and Alzheimer's disease [22]. Diverse drugs that are used in various psychological and neurological disorders might modify the GABA system at the level of the synthesis of GABA, induce anxiolysis or hypnosis in animals by potentiating the GABA-mediated postsynaptic inhibition through an allosteric modification of GABA receptors [23] and thirdly by direct increase in chloride conductance or indirectly by potentiating GABA-induced chloride conductance with simultaneous depression of voltage activated Ca++ currents like barbiturates.[24]

In this study, CNS depressant activity of HAETP was evaluated by rota rod test, which has clearly demonstrated the CNS depressant activity evidenced by decreased fall off time and another important activity of evaluating CNS drug action is to observe its effect on locomotor activity of the animal. The activity is a measure of the level of excitability of the CNS, and decreased activity results from CNS depression. The extract significantly decreased the locomotor activity as observed in the results of the actophotometer test. [Table 1&2] Moreover, anxiolysis was studied by open field method. It is used for evaluating the effect of drugs on gross general behavior and is used to measure the level of nervous excitability when the animals are exposed to a novel environment [25].

As above results, the HAETP possesses various phytochemical substances such as alcoholids, flavonoids, tannins, saponons, terpenoids and phytosterols. Several plants have been reported to have CNS depressant and anxiolytic activity due to the presence of triterpenoids, saponin and flavonoids. Triterpenoids, saponin and flavonoids are to have agonistic/facillitatory activities at GABA receptor complex, which led to the hypothesis that they act as benzodiazepine – like molecules. This is supported by their behavioural effects in animal models of CNS depressant and anxiety [26,27].

## 5. Conclusion

From the results we can conclude that HAETP possesses considerable CNS depressant and anxiolytic activity which is comparable with the standard through binding to benzodiazepines site on GABA-BDZ receptor complex. Nevertheless, further advance investigations are required to elucidate the exact mechanism involved in sedative effects and to identify the bio-active compound(s) associated with observed activity in animal behavioral models.

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