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Research Article

PHARMACOLOGICAL EVALUATION OF ANTI-DEPRESSANT ACTIVITY OF DESMOSTACHYA BIPINNATA LEAVES IN RATS

Vedamari Swathi* ¹, Murali Sollu ¹

¹ Department Of Pharmacology, KGR Institute Of Technology and Management, Medchal-Malkajgiri, Hyderabad – 501301, Telangana, India.

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Abstract:

Depression is a widely prevalent form of mental illnesses worldwide. It is commonly associated with sad mood, loss of interest or pleasure, feelings of guilt or low self-worth, disturbed sleep or appetite, and low energy. Desmostachya bipinnata has many medicinal properties, and are used in traditional medicine in the treatment of various medical conditions. This study was conducted to better understand the antidepressant activity of Desmostachya bipinnata. To evaluate the in vivo antidepressant activity of Ethanolic extract of Desmostachya bipinnata leave in Swiss albino mice. Ethanolic extract of Desmostachya bipinnata (EEDB) leaves was prepared by a continuous method using Soxhlet apparatus. The extract was subjected to phytochemical screening followed by acute oral toxicity studies in mice. EEDB in the doses of 100mg/Kg, 200mg/Kg and 400mg/Kg mg/kg body weight was administered to test groups Group 3, 4 and 5 respectively. Imipramine hydrochloride 15mg/kg body weight was administered to Standard group by oral route. Test group 3 received 100mg/kg (p.o). Control group received Normal saline 10ml/kg body weight. Antidepressant activity was identified by using modified Forced Swimming Test (FST) and Tail Suspension Test (TST). Period of immobility was observed in both the models which was indicative of anti depressant activity. Standard statistical methods were used to evaluate the results. The results showed significant dose dependent antidepressant effect of EASL in Swiss albino mice for both the models in all the test groups (Test group I, II and III). EEDB possess significant antidepressant activity. However, further investigations are required to determine its active constituents and molecular level of target mechanism of the extract for further use in humans.

Keywords: Desmostachya bipinnata, Antidepressant activity, forced swim test, Open Field Test.

Corresponding author:

Vedamari Swathi,

Department of Pharmacology,

Kgr Institute of Technology and Management,

Medchal-Malkajgiri, Hyderabad – 501301, Telangana, India,

Mail id: swathi.vedamari@gmail.com.

QR code

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INTRODUCTION:

Depression: It is basically acknowledged as illness with symptoms such as anxiety and sleep disturbances. It can be a persistent, recurring illness that can cause many personal suffering for individuals and their families. At present, disability caused by depression is estimated to be the fourth most important cause of worldwide loss of life years. This has resulted into a requirement of search for effective treatments, including antidepressant drugs, herbal remedies, psychotherapy and electroconvulsive shock therapy.

The Neurobiology And Pharmacology Of Depression:

I. Neurotransmitter Systems

Within the central nervous system (CNS), the catecholamines, adrenaline, noradrenaline and dopamine forms the adrenergic systems. Out of these, few of the adrenergic neurons are radiating from the ancient limbic system and plays to role of discharging

the catecholamines within the frontal cortex. Thus, the catecholaminergic pathways are claimed to be responsible for mood, alertness and stress responses. The primary neurotransmitter, which modulates the excitatory catecholamine systems of the CNS is Serotonin. The Serotonin neurons are responsible for the control of memory, mood, sex drive and appetite.

The systems of serotonin and noradrenaline are the important their main cell small bodies in brainstem areas that serve as headquarters for shipping axonal projections by the brains in specific pathways that mediate specific functions (See Figure No. 1 for an illustration of the serotonin projections for an illustration of the noradrenergic projections).

Multiple serotonergic and noradrenergic pathways may be dysfunctional in depression, generating many different symptoms.

Locus ceruleus projections

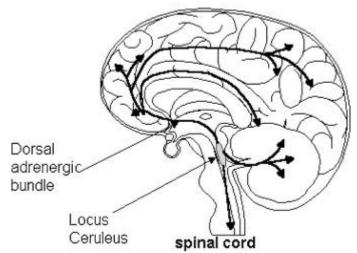


Figure No. 1: The projection of the noradrenaline system

The nucleus of the dorsal raphe projects the serotonin system and the raphemagnus. The serotonin receptors (5-HT) have been identified into various sub-types with the 5-HT1 and 5- HT2 sub-types being of greater interest in psychiatry. The most important of the 5-HT1 subclass is 5- HT1A which is concentrated in the hippocampus and raphe. The release of this 5 – HT from presynaptic neurons is modulated by this autoreceptor. The 5 - HT2 rceptors occur in high concentrations in the frontal cortex and nucleus accumbens. [2]

II. Hypotheses of Depression

Several hypotheses of the biological determinants of depression have emerged over the past century. The most important of these and the implications thereof are reviewed below. Today it is generally accepted that depression is not necessarily due to a shortage of one vital brain neurotransmitter, but rather to a disruption in the equilibrium between different regulatory systems.

A. The Biogenic Hypothesis of depression

The most common characteristic of depresion as claimed by monoaminergic hypothesis are a result of inadequate concentration of serotonin and noradrenaline in the synaptic clefts of the neurons in

the brain.³ This hypothesis has evolved to consider the possibility that depression may be the result of a deficiency in signal transduction from the monoamine neurotransmitter to its postsynaptic neuron, even with normal levels of neurotransmitter and receptor being present. Emerging theories that link genetic and environmental risk factors for depression suggest that stress can cause depression by downregulating certain genes, resulting in less key gene products, such as the brain-derived neurotrophic factor (BDNF), being produced. BDNF sustains the viability of neurons, so if the encoding gene is repressed the result may be atrophy or even apoptosis of neurons.

B. The dopamine hypothesis of depression

The original hypothesis was formulated in the late nineteen seventies by Solomon Snyder and linked schizophrenia with dopamine (DA) activity. Later, this hypothesis was extended to include depression following the observation that many antidepressants influence the metabolism of dopamine. Following chronic antidepressant treatment, the presynaptic DA receptors become subsensitised and this gets in an enhancement of DA release. A reduction in homovallinic acid (HVA), the main metabolite of dopamine, in the cerebral spinal fluid (CSF) of depressed patients who demonstrate marked motor retardation has also been reported. Therefore, a decrease in the ratio of HVA to DA is indicative of decreased turnover of DA. This hypothesis is also supported by reports of significantly reduced dopamine turnover in depressed suicide victims.

C. The permissive hypothesis of depression

This hypothesis emphasizes 5-HT as a neuro-modulator and its importance as a focus for antidepresant action. According to this theory, a lowered concentration in the central nervous system (CNS) of 5-HT results in an affective state regulated by NA. Decreased 5-HT and NA levels will give rise to depression. This Averages that 5-HT may act as a 'permissive' modulator of neurotransmitter function through connections between serotonergic pathways and make connections with noradrenergic and dopaminergic pathways via the associated receptors.

D. The glutamatergic N-methyl-D-aspartate hypothesis

As per recent researches, one of the important roles involved in the mechanism of depression is dysfunction of CNS glutamatergic pathways. Many of the researches confirm that the compounds, which induce reduction in the activities at the N-Methyl-D-Aspartate receptors produce effects similar to pharmacologically active antidepressants. Hence, it is

assumed that the common pathway affected by antidepressant drugs, whenever there are adaptive changes in NMDA receptor complex.

E. The kynurenine hypothesis

This hypothesis emerges from the premise that depression arises from altered levels of serotonine (5-Hydro. Trypt.) in the mind Serotonin is a metabolite of the esential A. A. tryphtophan (TRP) and all 5- Hydro. Trypt required by the neurons in the brain is synthesized in the brain because 5- Hydro. Trypt is unable to cross the BBB. Therefore, the availability of TRP is essential for the synthesis of 5-HT in CNS. There are several factors which affect the production and transport of TRP from the blood stream into the CNS, in which deficiency of Vit. B6, Stress, escalated cortisol levels and even high doses of TRP (2000m.g. of TRP). These are the factors simulating the conversion of TRP into kynurenine, which further results ito reduced TRP level. Therefore, the inhibition of liver enzyme tryptophan 2,3-dioxygenase (also known as tryptophan pyrrolase) during the first and rate - limiting step of the pathway of kynurenine would enhance circulating levels of TRP and thereby lead to increased neural production.

III. Treatments for Depression

MAOIs & tricyclic antidepresants (TCAs) were launched as the drug products approximately 60 years ago. These were found to have many side effects and to be highly toxic in the treatment of depression. This resulted into introduction of the selective noradrenaline reuptake inhibitors (SNRIs) and selective serotonin reuptake inhibitors (SSRIs), which are better tolerated and safer. However, these have not been shown to be conclusively superior to the TCAs and MAOIs. [4]

The chemical structures of antidepressant drugs vary significantly and therefore cannot be considered to be the most important factor in the search for new drugs with antidepressant activity. However, the mechanism of action of these drugs has provided insights into the pathology of depression. The basic biochemistry and possible Mode of action of major categories of antidepressant drugs are discussed below.

A. Tricyclic Antidepressants (TCA)

These drugs all have a characteristic three ring structure and are chemically similar to the phenothiazines. The discovery of their antidepressant action was fortuitous when imipramine, originally considered as a neuroleptic was found to have antidepressant activity.

Thereafter, first generation antidepressants emerged which display activity as mixed noradrenaline and serotonin reuptake inhibitors. The reuptake of monoamine neurotransmitters into the presynaptic neuron is inhibited by many of the TCAs by competitive inhibition of the ATPase in the membrane pump. Some TCAs are more selective than others but this has not been shown to influence the efficacy of the drug. The different monoamine reuptake properties can also include an increase in dopaminergic activity via a presynaptic mechanism for amitriptyline and a post synaptic mechanism for desipramine and imipramine.⁵

The major drawback of the TCA drugs is the side effects which resu lt from their antimuscarinic, antihistaminic and alpha adrenoceptor-blocking activity.

B. Selective Serotonin Reup take Inhibitors (SSRI)

Unlike the tricyclic antidepressants, the SSRIs reduce the neurona l uptake of serotonin but have no effect on noradrenaline. Therefore the SSRIs have a better side effect profile in comparis on with TCAs because these drugs have a low affinity for muscarinic, histaminergic and adrenergic receptors.

Fluoxetine was the first SSRI to be used clinically followed by paroxetine and sertraline. The latter two hav e shorter half lives and different potencies as inhibitors of specific P450 isoenzymes.⁶

C. Monoamine Oxidase Inhibitors (MAOI) The mechanism of action of MAOIs is complex, but their primary action is to inhibit the enzyme, monoamine oxidase (MAO), which is responsible for degrading free monoamines. There are two isoforms of MAO, designated MAO-A and MAO-B, with MAO-B being the predominan t in many parts of the brain. MAO inhibitor s have many side effects, but recently the reversible MAO-A inhibitor, moclobemide, was introduced which has fewer side effects. T oday, many people are searching for natural remedies to overcome depression. These have fewer side effects and are ea sily obtainable.

In general, depression is belongs to the group of most common psychological disorders. It is a heterogeneous disorder that has been characterized and classified in a variety of ways. Affective disorders are characterized primarily by change of mood (depression or mania) rather than by thought disturbance. Depression may vary from a mild condition to severe depression known as psychotic depression including delusions and hallucination.

The most commonly prescribed drug for depression such as imipramine, desipramine, fluoxetine etc. nowadays have been limited due to their potential side effects like anorexia, ant cholinergic effect (drug mouth, constipation, blurred vision, urinary retention), cardiovascular collapse, diarrhea, difficulty in concentrate, dizziness, drowsiness, dysphoria, dysrhythmias, epigestric distress, excessive central stimulation (excitement, convulsions), hypothermia, hypotension, insomnia, liver damage, loss of libido and failure of orgasm, mania, metallic taste, nausea, palpitation, postural hypotension, restlessness, sedation, seizures, tachycardia, tremor, weakness and fatigue, weight gain, and many other.6

MATERIALS AND METHODS:

The designing of methodology involves a series of steps taken in a systematic way in order to achieve the set goal (s) under the prescribed guidelines and recommendations. It includes in it all the steps from field trip to the observation including selection and collection of the medicinal plant, selection of dose value, standardization of protocol, usage of instruments, preparation of reagents, selection of specific solvents for extraction, formation of protocols and final execution of the standardized protocol. All this requires good build of mind and a good and soft technical hand to handle the materials and procedure in a true scientific manner.

Drugs and Chemicals:

Drugs and Chemicals used in this study were of analytical grade and of highest purity procured from standard commercial sources in India.

Table : Drugs and Chemicals

S.No Materials Company Name					
Materials	Company Name				
Mucrus	Company Name				
Iminramine	Drug Procured from Intas Pharma				
mipramine	Drug Procured from mas Pharma				
	Materials Imipramine				

Instruments

Following instruments were required for the study:

Table: List of Instruments used for study

Name of the instrument	Source				
Centrifuge	Dolphin				
Digital weighing balance	Horizon				
Glucometer	Horizon				
Heating mantle	ASGI®				
Refrigerator	Videocon				
Actophotometer	Dolphin				
Elevated Plus maze apparatus	Dolphin				
Glass cylinder	ASGI®				
Adhesive tape	YVR medivision Pvt Ltd				
Thread	YVR medivision Pvt Ltd				
Stop watch	ASGI®				
Syringes	YVR medivision Pvt Ltd				
Needles	YVR medivision Pvt Ltd				
Soxhlet extractor	ASGI®				
Condenser	ASGI®				
Burette stand	Dolphin				
Round bottom flask	ASGI®, Amar				
Mixer	Videocon				
Oven	ASGI®				
Water bath	ASGI®				
Stirrer/glass rod	ASGI®				
Watch glass	ASGI [®]				
Whatmann filter paper	Manipore microproducts				
	Ghaizabad.				
Butter paper	ASGI [®]				
Spatula	ASGI®				
Rubber pipes ASGI®					

Experimental animals:

Swiss albino rats, 60 in number, weighing 150-200 gms, of either sex, maintained under standard conditions in the Institutional animal house were used. They were housed in clean, transparent polypropylene cages in groups of six and maintained at standard laboratory temperature and humidity (40-60%) with light/dark cycle of 12:12 hours. Animals were fed commercial pelleted chow and water. The rats were allowed to acclimatize to these conditions for a week before starting the experiments. The standard drug, Imipramine hydrochloride, was obtained from Abbot Healthcare Pvt Ltd (Depsonil 25).

Wistar rats (150-200 g) and Swiss albino Rats (18-22g) of either sex selected for the study. Animals were housed in appropriate cages in uniform hygienic conditions and fed with standard pellet diet (Amrul Laboratory Animal Diet) and water ad libitum. All the animals were maintained under standard conditions, that is room temperature $26 \pm 1^{\circ}\text{C}$, relative humidity 45 - 55% and 12:12 h light – dark cycle. Animal studies had approval of IAEC.

Plant Material Collection

The aerial part of *Desmostachya bipinnata* was collected from Tirumala hills, Tirupati, Andhra Pradesh. India. It was identified and authenticated by Prof. Madhava Chetty, K., Taxonomist, S.V. University, Tirupati, Andhra Pradesh, India. A voucher specimen has been kept in our laboratory for future reference.

The plant material was cleaned, reduced to small fragments, air dried under shade at room temperature and coarsely powdered in a mixer. The powdered material was stored or taken up for extraction process.

Preparation of plant extracts: Preparation of Ethanolic Extract:

The *Desmostachya bipinnata* plants were washed, the leaves were shade dried and powdered. About 200 g of the dried leaf powder of *Desmostachya bipinnata* was extracted with 99.9% ethanol in Soxhlet extractor for about 36 hours. The ethanol was then evaporated from the mixture by placing it in a beaker and heating it over

a water bath. The extract gave a yield of brownish paste like mass weighing 6g. The yield obtained was 3% w/w with respect to dried powder.

Preliminary phytochemical screening:

Preliminary phytochemical screening of the plant extract was carried out for the analysis of Alkaloids, Carbohydrates, Tannins, Saponins, Steroids, Phenols, Flavonoids .as per the standard methods.

- **1. Detection of Alkaloids:** Extracts were dissolved individually in dilute Hydrochloric acid and filtered.
- a) Mayer's Test: Filtrates were treated with Mayer's reagent (Potassium Mercuric Iodide). Formation of a yellow coloured precipitate indicates the presence of alkaloids.
- **b).Wagner's Test:** Filtrates were treated with Wagner's reagent (Iodine in Potassium Iodide). Formation of brown/reddish precipitate indicates the presence of alkaloids.
- **c).Dragendroff's Test:** Filtrates were treated with Dragendroff's reagent (solution of Potassium Bismuth Iodide). Formation of red precipitate indicates the presence of alkaloids.
- **d).**Hager's Test: Filtrates were treated with Hager's reagent (saturated picric acid solution).Presence of alkaloids confirmed by the formation of yellow coloured precipitate.
- **2. Detection of Carbohydrates:** Extracts were dissolved individually in 5ml distilled water and filtered. The filtrates were used to test for the presence of carbohydrates.
- a).Molisch's Test: Filtrates were treated with 2 drops of alcoholic α -naphthol solution in a test tube. Formation of the violet ring at the junction indicates the presence of Carbohydrates.
- **b).Benedict's Test:** Filtrates were treated with Benedict's reagent and heated gently. Orange red precipitate indicates the presence of reducing sugars.
- **c).Fehling's Test:** Filtrates were hydrolysed with dil. HCl, neutralized with alkali and heated with Fehling's A&B solutions. Formation of red precipitate indicates the presence of reducing sugars.

3. Detection of saponins

- a). FrothTest: Extracts were diluted with distilled water to 20ml and this was shaken in a graduated cylinder for 15 minutes. Formation of 1cm layer off a am indicates the presence of saponins.
- **b).FoamTest:**0.5gm of extract was shaken with 2ml of water. If foam produced persists forten minutes it indicates the presence of saponins.

4.Detection of steroids.

a).Salkowski's Test: Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of Conc. Sulphuric acid, shaken and allowed

to stand. Appearance of golden yellow colour indicates the presence of triterpenes.

b).Libermann Burchard's test: Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of acetic anhydride, boiled and cooled. Conc. Sulphuric acid was added. Formation of brown ring at the junction indicates the presence of phytosterols.

5. Detection of Phenols

Ferric Chloride Test: Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenols.

6. Detection of Tannins

Gelatin Test: To the extract, 1 % gelatin solution containing sodium chloride was added. Formation of white precipitate indicates the presence of tannins.

7. Detection of Flavonoids

Alkaline Reagent Test: Extracts were treated with few drops of sodium hydroxide solution. Formation of intense yellow colour, which becomes colourless on addition of dilute acid, indicates the presence of flavonoids.

Leadacetate Test: Extracts were treated with few drops of lead acetate solution. Formation of yellow colour precipitate indicates the presence of flavonoids.

Selection of dose for animal study

The dose considered for the experiment on rats was obtained from conversion of human dose of *Desmostachya bipinnata* (3-5 g/kg). The conversion factor of human dose (per 200 g body weight) is 0.018 for rats and 0.002 for Rats (Ghosh 1984). Hence the calculated dose for the rats (considering human dose 3 and 5 g/kg) is 200 mg/kg and for Rats is 20 mg/kg. Acute toxicity was done at dose of 2000mg/kg body weight.

Pharmacological evaluation: Acute oral toxicity:

The acute oral toxicity Ethanolic extracts of *Desmostachya bipinnata* was determined by using rats which were maintained under standard conditions. The animals were fasted 12 hour prior to the experiment, up and down procedure OECD guideline no. 425 were adopted for toxicity studies. Animals were administered with single dose of individual extract up to 2000mg/kg and observed for its mortality during 2days and 7days study period (short term) toxicity and observed up to 7days for their mortality, behavioral and neurological profiles.

Screening for antidepressant activity:

The Ethanolic extracts of *Desmostachya bipinnata* leaves were tested for antidepressant activity using despair swim test and tail suspension test.

Treatment

The Wistar albino rats (n=60) were divided into two arms which was further divided into five groups, each group having six Wistar albino rats. Drugs were given orally after 12 hours of fasting every day, for ten days. The drugs were prepared and administered per oral (0.1 ml/10 g).

Group 1 was administered normal saline (10ml/kg). Group 2 was given standard drug Imipramine (15mg/kg). 12

Group 3, 4 and 5 received 100mg/kg, 200mg/kg, and 400mg/kg doses of the test compound Ethanolic Extract of *Desmostachya bipinnata* respectively.

For the Acute study, on day 1, one arm of 30 Wistar albino rats were subjected to Tail Suspension Test (TST), while 30 rats in the other arm were subjected to Forced Swim Test (FST), one hour after feeding the respective drugs. For Sub acute study, on day 10, the Wistar albino rats were again subjected to TST and FST, one hour after feeding respective drugs.

Procedure for antidepressant activity: Forced swim test (fst):

The method used was as described by Porsolt et al. The rats were individually forced to swim in a vertical plexiglass cylinder (capacity: 5L, height: 50cm diameter: 18cm) containing 15cm of water maintained at temperature: 25°C. Rats were subjected to prescreening, which lasted for 15 minutes. 24 hours after pre-screening, the trial was performed for 6 minutes of which the first two minutes were not recorded, and the periods of immobility for the latter four minutes was measured (in seconds) with a stopwatch. Rats were considered to be immobile when they made only the bare necessary movements to stay afloat, or when they were motionless. The Rats were taken out of the plexiglass cylinder after 6 minutes. They were dried with a dry towel, and kept under a dim lamp for drying. The water was discarded after every test, and fresh water was used for the next rats.

Tail suspension test (tst):

The method used was as described by Steru et al. Antidepressants that are used in practice are able to reduce the period of immobility of rats when they try to escape when suspended by their tail. This test was a reliable screening method for antidepressants, including those involving serotonergic system. Rats

ware hung on a wooden rod, 50 cm above the table, by attaching them from their tail end with the use of an adhesive tape. The first two minutes were not recorded, and the periods of immobility for the latter six minutes was recorded (in seconds) with a stopwatch. Rats were considered to be immobile only when they were motionless and not attempting to

Open-field test:

For open-field test, animals were divided into four groups (n = 10 / group): control (0.9% saline), the three doses of Desmostachya bipinnata (100, 200, 300 mg/kg) for one-week treatment. To assess the effect of Desmostachya bipinnata on locomotor activity, Rats were evaluated in the open-field paradigm (TRU SCAN Activity Monitoring Systems, Coul bourn Instruments) previously described. Animals were individually placed in a box $(40 \times 60 \times 50 \text{ cm})$. The rats were not habituated to the box before the test. The Rats were placed in the center and their behavior was noted immediately and continued for 4 min. The parameters such as total movements, total distance, total ambulatory move time were recorded by video camera and registered in the computer. During the interval of the test the apparatus was cleaned.

Statistical analysis:

Statistics The recorded data was entered in Microsoft Excel. The variables recorded followed normal distribution, hence, results have been expressed as mean (in seconds) \pm standard error of mean (SEM). The data was analysed using one way ANOVA followed by post-hoc Dunnet's test. Probability 'p' value less than 0.05 was considered as statistically significant.

RESULTS:

Phytochemical screening test

The freshly prepared extract of the leaves of Desmostachya bipinnata was subjected phytochemical screening tests for the detection of various active constituents. The extract showed the presence of alkaloids, tannins, steroids, phenolic and flavonoids, carbohydrates, and glycosides in crude extract of Desmostachya bipinnata leaves as depicted in Table 1.

Table: Result of chemical group tests of the Ethanolic extract of Desmostachya bipinnata leaves.

Carbohydrates	++
Tannins	+
Flavonoid	++
Saponin	+
Phenols	++
Steroids	+
Alkaloids	++
Glycosides	+

Antidepressant activity of desmostachya bipinnata:

In the Acute study, on Day 1, standard drug Imipramine (15mg/kg) and test drug EEDB (100mg/kg, 200mg/kg, 400mg/kg) showed significant reduction in immobility times when compared to control in both FST and TST (Table 1, Figure 1). In the Sub acute study, on Day 10, both Imipramine (15mg/kg) and EEDB (100mg/kg, 400mg/kg) showed significant reduction in immobility times when compared to control in both FST and TST.

Table: Effect of extracts of Desmostachya bipinnata on Anti-depressant activity

Day 1	Tail Suspension Test (Time in Sec)	Forced Swim Test
Normal Saline	225.1(±02.04)	139.2(±1.19)
Imipramine 15mg/kg	176.0 (±15.03)*	103.21(±0.14)*
EEDB 100mg/Kg	163.2 (±16.2)*	98.01(±2.01)*
EEDB 200mg/kg	183.1 (±6.10)*	110.10(±1.06)*
EEDB 400mg/kg	118.3 (±10.06)*	108.06(±2.19)*

Immobility time shown in seconds as mean $(\pm$ SEM),*denotes statistically significant value, # denotes statistically not significant value.

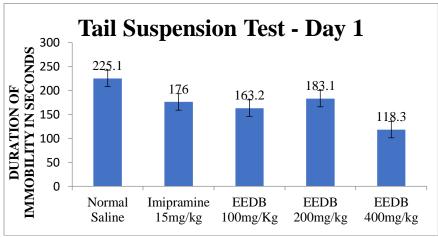


Fig: Tail Suspension Test Day-1

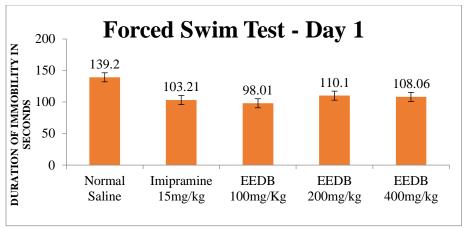


Fig: Forced Swim Test Day-1

Table: Effect of extracts of Desmostachya bipinnata on Anti-depressant activity

Day 10	Tail Suspension Test	Forced Swim Test	
Normal Saline	$225.25(\pm 12.08)$	142(±1.26)	
Imipramine 15mg/kg	148.61(±29.41)*	95.25(±2.95)*	
EEDB 100mg/Kg	95.98(±3.152)*	70.50(±3.24)*	
EEDB 200mg/kg	210.16(±10.76)#	95.19(±9.16)*	
EEDB 400mg/kg	105(±12.61)*	89.39 (±01.28)*	

Immobility time shown in seconds as mean (±SEM), *denotes statistically significant value, #denotes statistically not significant value.

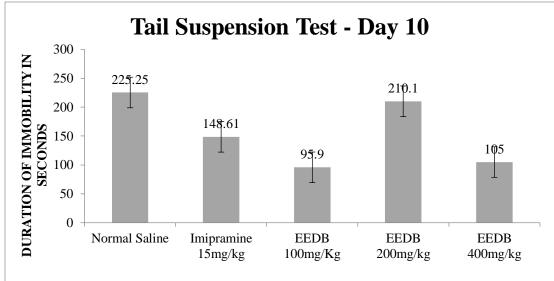


Fig: Tail Suspension Test Day-10

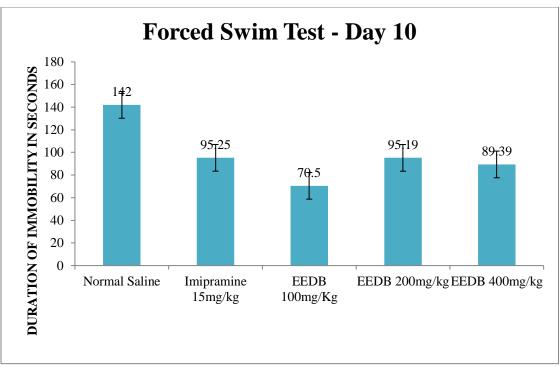


Fig: Forced Swim Test Day-10

Open field test:

Antidepressant activity of Ethanolic solvent soluble fraction of the leaves of *Desmostachya bipinnata* studied at a dose of 200 mg/Kg, using Forced Swim Test experiment.

In tail suspension test, Ethanolic extracts of leaves of *Desmostachya bipinnata* at a dose of 200 mg/kg i.p. significantly decreased the immobility time. The magnitude of the antidepressant effects of 200 mg/kg i.p. of alcoholic leaves of *Desmostachya bipinnata* was comparable to that of Imipramine 15mg/kg i.p. (Table -4).

Table : Effect of Ethanolic Extracts of *Desmostachya bipinnata* Leaves on Open Field TEST in Rats at Different Time Intervals

S.No Treatment		Dose (mg/kg)	Duration of immobility		%change in activity	
5.140	5.No Heatment	Dose (mg/kg)	Before	After	70 Change in activity	
1.	Control		39			
2.	Standard	15	22	122	85.31%	
3.	EEDB	200	43	183	76.9%	
4.	EEDB	400	56	165	65.6%	

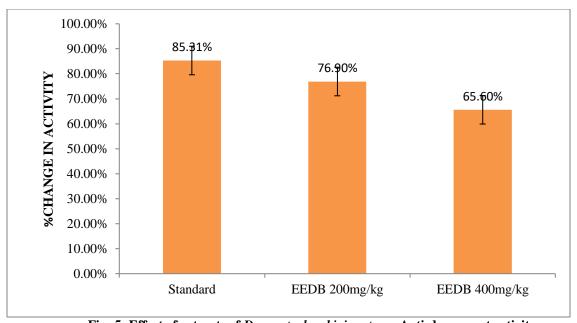


Fig -5: Effect of extracts of *Desmostachya bipinnata* on Anti-depressant activity
Table 5: Effects of *Desmostachya bipinnata* on duration of immobility time in open field test (OFT)

Treatments	Dose (mg/kg)	TL	CL	PL	L	G	D
Control		140.1±3.1	30.1±6	110.1±5.14	8.0±1	1.5±1.51	0.31±1.00
Diazepam	10	152.6±2.72	36±2.08	116.6±2.33	9.0±1.52	0.33±0.32	0.0±0.0
EEDB	200	119.3±5.24	24.3±2.4	95.0±5.5	11.3±2.02	1.33±0.32	0.0±0.0
EEDB	400	135.6±13.5	27.3±5.04	27.3±5.04	10.3±1.20	2.33±1.20	0.0±0.0

Values are expressed as Mean \pm S.E.M (n=10). *P<0.05, **P<0.01, ***P<0.001 when compared with control groups. TL: Total Locomotion, PL: Peripheral Locomotion, CL: Central Locomotion (CL), L: leaning, G: grooming, D: defectation.

For the open field test number of line crosses and the frequency of rearing are usually used as measures of locomotor activity, but are also measures of exploration and anxiety. A high frequency of these behaviors indicates increased locomotion and exploration and/or a lower level of anxiety. The number of central square entries and the duration of time spent in the central square are measures of exploratory behavior and anxiety. A high frequency/duration of these behaviors indicates high exploratory behavior and low anxiety levels.

DISCUSSION:

A previous study concluded that aqueous leaf extract of *Desmostachya bipinnata* exhibited antidepressant in forced swimming (FST) and tail suspension (TST) tests.

In this study, both Imipramine and *Desmostachya bipinnata* showed a reduction in immobility times in acute and sub acute study in both FST and TST. Lowest immobility times were recorded with *Desmostachya bipinnata* at 100 mg/kg doses in most recordings, and at times, it showed comparable or even better reduction in immobility times than Imipramine in both tests in acute and sub acute study.

Imipramine inhibits nor epinephrine transporter and Serotonin transporters, increasing their availability at synaptic cleft, thereby reducing depression. The antidepressant action of *Desmostachya bipinnata* is probably similar to the mechanisms of anti-depressant agents, like Imipramine, that are effective in the above screening models. Phytochemical investigations done in a study showed the presence of alkaloids, flavonoids and tannins in the extract. It is likely that the antidepressant activity seen with *Desmostachya*

bipinnata could be because of the above mentioned Phytoconstituents.

SUMMARY:

The fresh leaves of *Desmostachya bipinnata* used for this project work. The dried leaves of *Desmostachya bipinnata* were successively extracted with ethanol. Therapeutic dose of the extracts were calculated after carrying acute oral toxicity studies rats. Extracts were tested for their anti-depressant activity in Rats. By Forced Swim Test and Open Field Test.

On the basis of the clinical association of depressive episodes and stressful life events, many of the animal models for the evaluation of antidepressant drug activity assess stress precipitated behaviors. The Force Swim Test and Tail Suspension Test are the most commonly used animal models for anti depressant activity.

In the present study, forced swim stress (FST) and Tail Suspension was used to developed depressive like behavior in rats for 1 and 10 days. It was an increased immobility time, anxiety and impaired cognitive behavior in the experiment rats (stressed).

The present study provides the evidence indicating that Ethanolic extract of *Desmostachya bipinnata* showed significant antidepressant activity in TST and FST models of depression in Rats. The antidepressant activity may due to presence of phenolic components.

CONCLUSION:

In the present study plant parts of *Desmostachya bipinnata* have been be evaluated for antidepressant activity. As literature shows that traditionally this plant is being use in the treatment of depression. The plants materials *Desmostachya bipinnata* used for the present studies were commercially procured from local market. Albino rats were used for the antidepressant activity.

The present study provides the evidence indicating that methanolic extract of *Desmostachya bipinnata* showed significant antidepressant activity in TST and FST models of depression.

Phytochemical analysis showed the presence of Flavonoids and phenolic compounds have been reported to have multiple biological effects such as Central nervous system disorders.

Similarly, the results of this study suggest that the leaf extract exhibited significant antidepressant activity with a strong psychomotor stimulation. The leaf extract was reported to contain chemical constituents such as Carbohydrates, Tannins, Flavonoid, Saponins, Phenols, Steroids, Alkaloids and Glycosides.

The results obtained in this study suggest that Ethanolic Extract of *Desmostachya bipinnata* has antidepressant activity and can be considered for use in therapy of depression after further testing.

Further research is required to know the mechanism of its action.

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