



ISSN : 2347 - 2243

*Indo - American Journal of
Life Sciences and Biotechnology*



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Neuropharmacological Profiling of *Acanthus ilicifolius*: A Coastal Mangrove's Potential in CNS Depression and Analgesia"

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Abstract

An exceptionally abundant supply of significant chemicals with structurally unique and physiologically active metabolites may be found in the marine environment. Mangrove plants have long been utilized in traditional medicine to cure a variety of illnesses all around the globe. Very few efforts have been undertaken to test the validity of these claims in controlled trials, despite the fact that several mangroves and mangal companions are suggested in traditional medicine as being effective against a variety of ailments. *Acanthus ilicifolius* Linn, often known as sea holly (acanthaceae), is a salty plant that is typically found around the southeast coastlines of India. It is well-known for its wide range of secondary metabolites and its traditional uses. Using a Soxhlet apparatus, the leaves of *Acanthus ilicifolius* were gathered, shade-dried, ground into a coarse powder, defatted with petroleum ether, and then extracted with ethanol. Assessment of the effects of an ethanolic extract of *Acanthus ilicifolius* leaves as a central nervous system depressant. Mice were subjected to a number of behavioral tests, including the Tail Suspension Test (TST) and the Forced Swimming Test (FST). The immobility periods in the FST extract at dosages of 100, 250, and 500 mg/kg were considerably ($P < 0.0001$) longer than those in the control group. When compared to a normal medication, TST considerably lengthens the immobility period when the dosage is increased. The drug's interaction with the GABA ergic system may be the cause of these depressed patterns.

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Introduction

All plant components, including flowers, roots, stems, leaves, fruits, seed bark, and whole plants, have therapeutic qualities. Nevertheless, it has been noted that some plants are unsafe due to the presence of harmful substances that have negative physiological consequences. Around the globe, herbal medicine is extensively used. According to the World Health Organization (WHO), 4 billion people, or 80% of the global population, utilize herbal medicine for basic healthcare in one way or another. About 11% of the 252 medications are made from plants, and the WHO has acknowledged herbal medicine as a crucial part of basic healthcare [1]. In tropical and subtropical regions, mangroves make up the interface ecosystem that exists where rivers and the sea meet. Both the ecosystem and the plant groups that have adapted to thrive in the tidal saline environment are referred to as "mangroves" [2]. Extracts from mangrove plants have been used for generations to cure medical conditions. Due to their many uses, chemicals originating from plants have lately attracted a lot of attention [3]. Although the mechanisms of action of various kinds of CNS depressants vary, they are all capable of decreasing central nervous system activity and lowering brain consciousness. CNS depressants are used for conditions including insomnia, anxiety, epilepsy, stress before to surgery, and panic attacks.

Insomnia

The word "insomnia" refers to a group of issues including a persistent difficulty to get enough sleep. Up to 40% of the general population suffers from insomnia each year, and it is a major source of illness and death [4]. An imbalance between the neurotransmitters that promote sleep, Gamma-Amino Butyric Acid (GABA), leads to insomnia.

Anxiety

People with anxiety problems may also have other co-morbidities, such as mental health disorders, sleep issues, irritable bowel syndrome, and other illnesses [29]. Recently, neurochemicals including serotonin, GABA, dopamine, norepinephrin, and many more have been linked to anxiety disorders. Each molecule has a unique but equally important role in anxiety regulation. It is believed that noradrenergic system dysregulation is the root cause of anxiety disorders. Noradrenaline regulates autonomic arousal activities, including increased breathing and heart rate. The ensuing physiological cascade results in panic symptoms such as paraesthesia, numbness, and tightness in the chest. Generalized anxiety disorder (GAD) is associated with noradrenergic hyperactivity, dysregulation of the serotonin receptor (5-HT_{1A}, 5-HT_{2C}), and a decrease in the number of benzodiazepine sites on the GABA-benzodiazepine receptor complex [5].

Epilepsy: The disorder known as "epilepsy" is characterized by frequent, spontaneous seizures. There are several different causes of epilepsy, many of which point to underlying brain malfunction. With a frequency of around 50 new cases per 100,000 people annually, epilepsy is one of the most prevalent neurologic disorders. Roughly one-third of patients have refractory epilepsy, and approximately 1% of the population has epilepsy [6]. One way to think about a seizure is as a disruption of the brain's usual equilibrium between excitation (E) and inhibition (I). Many different levels of brain function may change, leading to an E/I imbalance. E/I balance may be affected by acquired or inherited causes. From circuit-level abnormalities (such as aberrant synaptic connectivity in cortical dysplasia) to receptor-level abnormalities (such as aberrant γ -aminobutyric acid [GABA] receptor subunits in Angelman syndrome) to ionic channel dysfunction (such as potassium channel mutations in benign familial neonatal epilepsy [BFNE]), genetic pathologies can result in epilepsy [6].

Materials and Methods Animals

For this investigation, male Swiss albino mice weighing 20–25g were used. In addition to being fed a regular pellet diet and given unlimited access to water, the animals were kept in standard ambient conditions ($25 \pm 2^\circ\text{C}$) and relative humidity (45 to 55%). They were also subjected to a 12-hour light and 12-hour dark cycle [7].

Plant Material

Mangroves and *Acanthus ilicifolius* Linn. (Acanthaceae) are prevalent in India's southeast shores, including Thiruvallur, Chennai, Chengalpattu, Pichavaram, Thiruvarur, and Nagapattinam. Using a Soxhlet apparatus, the



leaves of *Acanthus ilicifolius* were gathered, shade-dried, ground into a coarse powder, defatted with petroleum ether, and then extracted with ethanol.

Methods

Screening of CNS Depressant

1. Forced Swimming Test (FST)
2. Tail Suspension Test (TST)

Animals: Albino Mice

The animals were divided into five groups of six mice each.

Group I - Control group (distilled water with 2% DMSO)

Group II- is the standard group, which receives 5 mg/kg of diazepam. **Group III**: Test (100 mg/kg of ethanol extract in 2% DMSO)

Group IV - Test (Ethanol extract in 2% DMSO at the dose of 250mg/kg)

Group V - Test (Ethanol extract in 2% DMSO at the dose of 500mg/kg)

Forced Swimming Test Procedure

The most popular behavioral paradigm for determining if rats are experiencing CNS depressive activity is the Forced Swim Test (FST). Each mouse was made to swim alone in a 25 x 15 x 25 cm glass container filled with fresh water up to 15 cm high and kept at $26 \pm 1^\circ\text{C}$. The water was so high that the animals could not use their tails or rear paws to sustain themselves by contacting the chamber's side walls or floor. After each animal was subjected, the water in the chamber was replaced. For the first two minutes of the test, every animal moved vigorously. The remaining four minutes of the six-minute testing session were spent manually recording the length of immobility.

When mice stopped struggling and stayed still in the water, it was thought that they were immovable and merely needed to lift their head above the water. Mice were dried with towels after swimming and put back in their home.

Tail Suspension Test ^[8]

Procedure

With the use of sticky tape positioned about 1 centimeter from the tip of the tail, mice were hung on the edge of the table, 50 cm above the floor. Six minutes were spent observing the length of immobility. During the last four minutes of the observation period, the length of immobility was noted. Only when mice hung passively and remained absolutely still were they deemed immobile. When the animal hung quietly and showed no movement of the body other than that necessary for breathing, it was deemed motionless. Animals were individually trained for 30 minutes and in 15-minute sessions prior to treatment. For six minutes, each animal was put through the Tail Suspension Test in a comparable setting, and the amount of time each animal remained immobile was noted.

Statistical Analysis

The mean \pm standard error of the animals in each group was used to represent all of the CNS depressant activity data. One-way ANOVA and Turkey's t-test were used for the statistical analysis. The ANOVA statistical program (Graph Pad and PRISM software version 9.0.1 (151)) was used for all analyses.

Results and Discussion Results

Forced Swimming Test

When compared to the control group, the extract at dosages of 100, 250, and 500 mg/kg significantly lengthened the immobility period (Fig. No. 1). Likewise, the duration of immobility was significantly increased by the conventional medication diazepam (5 mg/kg, i.p.).

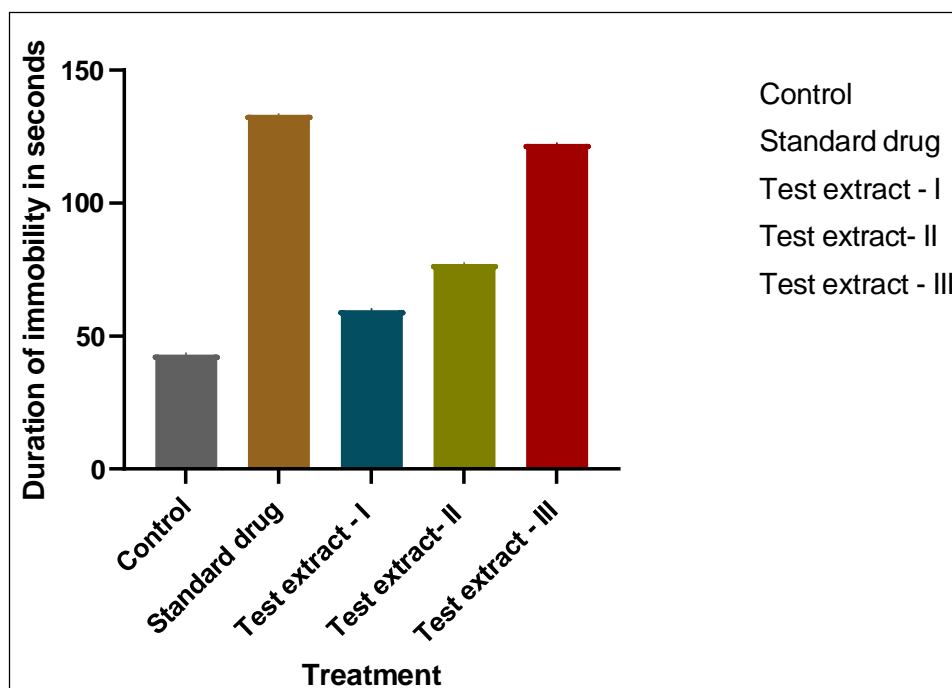
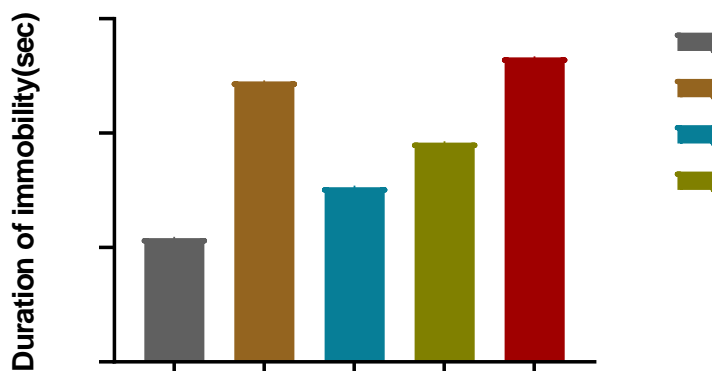


Fig 1: Effect of *Acanthus ilicifolius* extracts at different doses in Forced swimming test

Test extract I- 100mg/kg Test extract II- 250mg/kg Test extract III- 500mg/kg
Standard drug (diazepam) - 5mg/kg



Tail Suspension Test

The results for CNS depressant activity of test extracts in Tail suspension test are given in Fig. No. 2.

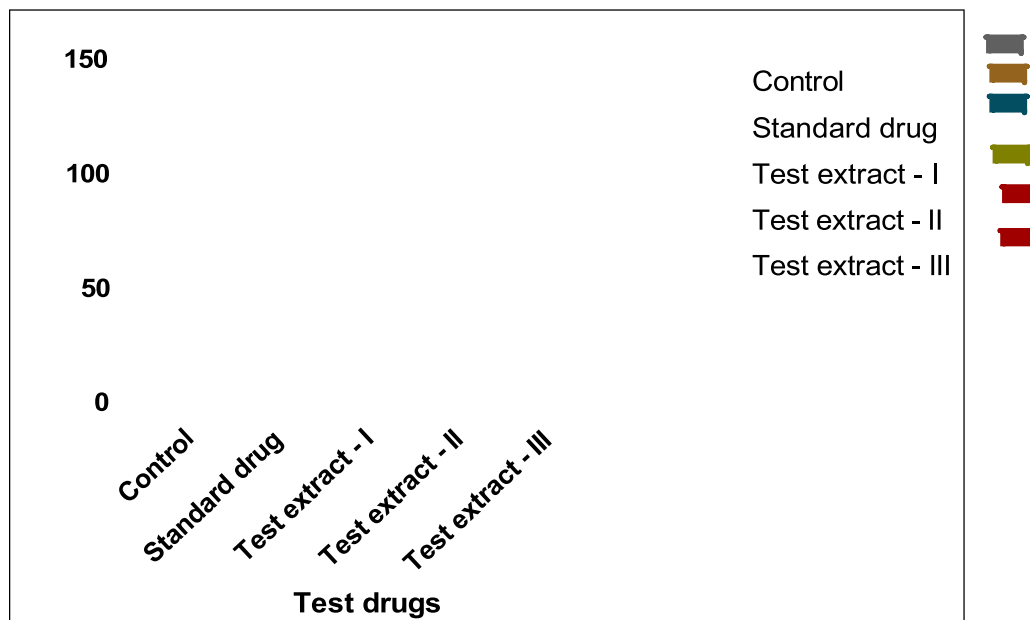


Fig 2: Effect of *Acanthus ilicifolius* extracts at different doses in Tail suspension test

Test extract I- 100mg/kg Test extract II - 250mg/kg Test extract III- 500mg/kg
Standard drug (diazepam) - 5mg/kg

Discussion

In comparison to the usual medication diazepam (5 mg/kg, i.p.) treated group, treatment with an ethanolic extract of *Acanthus ilicifolius* leaves at several dosages (100 mg/kg, 250 mg/kg, and 500 mg/kg p.o.) demonstrated a substantial central nervous system depressive effect in FST. The action of the test medication on inhibitory receptors found in the central nervous system may be the origin of the notable depressed pattern seen during the forced swimming test. [9] The frequency of CL- is increased by the modulatory receptors.

GABA submaximal concentration causes channel opening. decreasing the pace at which neurons fire and causing hyperpolarization as a result of cl-ion inflow [88]. The evaluation of EELAI in mice using the tail suspension test revealed an increase in the duration of immobility. Its dose-dependent CNS depressing impact was supported by the findings, which are summarized in Fig. No. 2. The test drug's binding to the GABA inhibitory receptor may be the cause of the result [9].

Conclusion

The traditional usage of *Acanthus ilicifolius*, a mangrove plant, as a central nervous system depressant has been well validated by the current investigation. The test compound increased immobility time in the forced swimming test by the same amount as a conventional drug. The test drug's inhibitory action on inhibitory receptors in the



central nervous system may be the origin of the notable depressed pattern seen in the forced swimming test. The tail suspension test likewise generated a dose-dependent depressive effect via interacting with the GABAergic system.

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