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ASSESSMENT OF PHYTOCHEMICAL COMPOSITION & PHARMACOLOGICAL INVESTIGATION *OF ECHINOPUS ECHINATUS* ROXB FLOWERS

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ABSTRACT

This research paper presents a comprehensive analysis of the phytochemical composition and pharmacological potential of *Echinopus echinatus* Roxb flowers, an indigenous plant with therapeutic relevance. The phytochemical screening revealed the presence of diverse secondary metabolites, such as alkaloids, flavonoids, phenolic compounds, and terpenoids, suggesting a rich chemical profile. Subsequently, the pharmacological investigation was conducted to evaluate the potential therapeutic effects of *Echinopus echinatus* Roxb flowers. In vitro assays were employed to assess antimicrobial activities. Additionally, in vivo experiments were conducted to evaluate the plant's anti-anxiety and anti-diabetic properties. The findings demonstrated significant anti-anxiety activities, suggesting the potential of *Echinopus echinatus* Roxb flowers in mitigating oxidative stress and inflammatory conditions. Overall, this study contributes valuable insights into the pharmacological properties of *Echinopus echinatus* Roxb flowers, shedding light on its potential applications in medicine and providing a scientific basis for further exploration and development of pharmaceutical products derived from this plant.

KEYWORDS: *Echinopus echinatus*, Anti-anxiety Drug, Phytochemical Screening, Herbal Anti-anxiety drugs.

I. INTRODUCTION

Throughout history, humans have utilized herbs and remedies to combat illnesses. The Indian system of medicine has a rich history deeply intertwined with our cultural heritage, catering to the healthcare needs of a significant portion of our population. This system mainly herbs. Herbal medicines, also known as Herbalism or Botanical medicine, consist of plants or plant parts appreciated for their medicinal properties. These herbs contain a range of chemical substances that have effects on the body. Herbalists utilize all parts of plants – leaves, flowers, stems, berries, and roots – to prevent, alleviate, and treat various illnesses. From the "scientific" perspective, many herbal treatments are considered experimental. If we seen in our past the Rugveda, one of the oldest repositories of human knowledge, mention the used of 67 plants for therapeutic use, they Yajurveda enlist 81 plants whereas the Atharvaveda written during 1200 BC describes 290 medicinal plants of medicinal value. Charak Samhita written during 990 BC describes 341 medicinal plants. The Ayurvedic milestone, the Sushrut Samhita, written around 600 BC, documented 395 medicinal plants. Subsequently, the Dhanwantari Nighantu listed 750 medicinal plants, with 450 mentioned in Bhavaprakash, 480 in Madanpala Nighantu, and 450 in the Kiiyadeva Nighantu. India leads in the utilization of herbal remedies. The prevalence of herbal drugs in India can be attributed to the country's rich biodiversity, abundant flora, and diverse geographical conditions, which support the cultivation of various exotic medicinal plants.[1] A systemic study of a crude drug under Pharmacognostic scheme its description on official title synonyms or vernacular names, biological source and family, geographical source or habitat history and introduction of crude drug. Also scheme on chemical constituent and qualitative chemical tests. A crude drug may have official or unofficial status. Unless otherwise indicates the official title by which it appears in the pharmacopeia books. Official title of a crude drug are given both English and Latin. The history of a crude drug reveals information about its information about its mankind. Evaluation of crude drugs means its identification and determination of its purity and quality. All this study are involved in pharmacognosy related to herbal plants.[2]

Introduction to *Echinops echinatus* Roxb.

Echinops echinatus Roxb, a member of the Asteraceae family, is a hairy annual herb reaching a height of 1-3 feet, with branches that spread widely from the base. The species is found practically throughout India, Pakistan, Afghanistan, etc.[3] In India it occurs in several areas such as Andhra Pradesh, Bihar, Himachal Pradesh, Jammu and Kashmir, Karnataka, Madhya Pradesh, Maharashtra, Rajasthan, Tamil Nadu, Uttar Pradesh, West Bengal.[4-6] Echinopes

echinatus an erect branched herb about a meter high. It features brief, sturdy stems that branch out from the bottom, adorned with white cottony hair. A stiff, young, yearly plant reaching heights of 1-3 feet, with branches spreading widely from the base. Its leaves are alternate, oblong, pinnatifid, and have a cottony underside, with triangular and oblong lobes that are spiny and often reach 2.5 cm in length. The deeply pinnatifid leaves are 7-12 cm long. The flower heads are white or purple, compact, and clustered at the branch ends; the involucres are surrounded by sturdy white bristles resembling pappus-hairs, with a short, yellowish pappus forming a small cylindrical brush above the achene. Heads occur in solitary white spherical balls, 3-5 cm across. Petals of the tiny white flowers are 5 mm long. Flowering periods occurs from December-January.[7] The plant is known for its bitter taste, appetite-boosting properties, and its ability to stimulate the liver. It is commonly employed in treating brain disorders, joint pain, and inflammation. In indigenous medical practices, the roots and root bark of the plant are utilized to address a range of health issues. Additionally, the root is used as both an abortifacient and an aphrodisiac.[8] The traditional healers of Chhattisgarh in India use this herbin different ways both internally and externally for the treatment of sexual disorders also.

Fig. 1: Flowers of *Echinopus echinatus* Roxb. Plant.

Scientific Classification[9]

Table No. 1: Scientific Classification.

Kingdom	Plantae
Division	Mangoliophyta
Class	Mangoliopsida
Subclass	Asteridae
Order	Asterales
Family	Asteraceae
Genus	*Echinops*
Species	*echinatus*

Morphological Characters

Flower: White or purple flower heads are compact, globose, and clustered at the ends of branches. Involucres are surrounded by robust white bristles that resemble pappus-hairs, with a short, yellowish pappus forming a cylindrical brush above the achene. The heads appear as solitary white spherical balls, measuring 3-5 cm across. The petals of the small white flowers are 5 mm long.[7]

Fig No. 2: Flower of Echinopus *echinatus* Roxb. Plant.

Chemical constituents in Flowers: Apigenin, apigenin 7-O-glucoside, and a new acyl flavone glucoside named echitin (I) were isolated from *Echinops echinatus* flowers. A minor alkaloid 7-hydroxyechinozolinone (I) is reported from the flowers of E.echinatus. Isomeric acyl flavone glycosides echinacin (I) and echinaticin (II) are reported from E. echinatus. Chaudhuri PK26 (1997) isolated Echinozolinone,[9] an alkaloid from *Echinopsechinatus.* Chaudhuri PK26 (1997) isolated Echinozolinone[10] an alkaloid from *Echinopsechinatus*. In addition to echinopsine and echinopsidine, a new alkaloid, echinozolinone, has been identified in *Echinopsechinatus*as 3(2-hydroxyethyl)-4(3H)-quinazolinone.

II. MATERIAL AND METHODS

Plant collection and authentication

The flowers of ***Echinopus echinatus*** tree were collected from botanical garden of S.R.T.M. krishividyapith, Nanded. The plant authenticated in department of botany PDEA's Waghire College, Saswad, Tal- Purandar, Dist - Pune. It is were dried under shade and made into coarse powdered by mechanical process and stored in air tight container for further study.

Chemicals and drug: All organic solvent and chemicals were purchased from S.D. fine chemicals Ltd; Mumbai and were of analytical grade.

Animals: The Albino mice 25-30 mg of either sex are obtained from S.G.R.S. College of Pharmacy, Saswad, Pune.

Phytochemical extraction: Extraction was carried out by Soxhlet (Continuous successive) extraction.

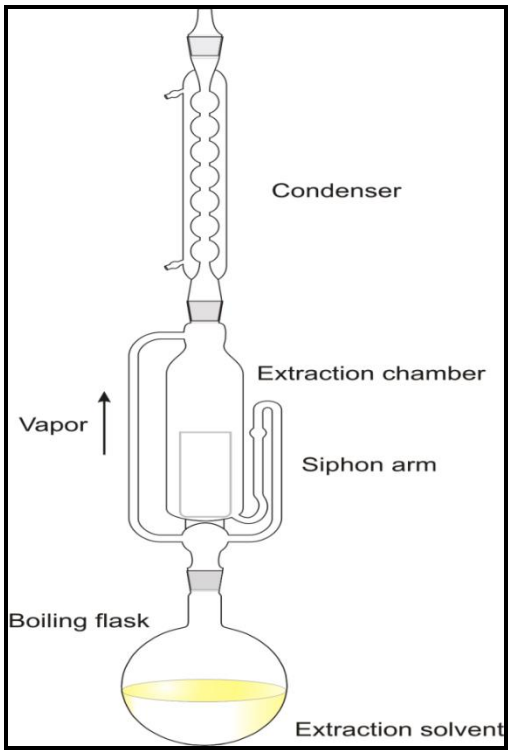

Fig. No. 3: Soxhlet Extractor.

Soxhlet extraction: The 20gm of coarsely dried powder of ***Echinopus echinatus*** flower were subjected to continuous hot extraction with various solvents separately. The extracted were filter and dried. The crude extract analyse the presence of various phytoconstituents by following standard phytochemical test.

Pharmacological Evaluation

To perform CNS depressant Activity of *Echinopus echinatus* flower

The various methods used,

1. Locomotar activity.
2. Antianxiety activity.
3. Phenobarbitone induced sleeping time.

1. **Locomotar Activity: By using Actophotometer**

Treatment Schedule

1. Group1-Distilled water containing tween80 o.p.(control).
2. Group2-Diazepam (2mg/kg) orally.(standard).
3. Group3-methanolic extract. o. p.
4. Group4- chloroform extract. o.p.
5. Group5-Aquoeus extract. i.p.

Animal Albino Mice(22-30gm) of either sex used in this experiments Animal where provided with standard food and water *ad libitum* and were maintained at temperature 25±2°C, humidity of 55±5% and with 12 hr. light - dark cycle. All animal procedures have been approved and prior permission from the Institutional Animal Ethical Committee was obtained as per the prescribed guidelines.

Fig. 4: Albino Mice.

III. RESULTS AND DISCUSSION

Extraction: The flowers of plant shade dried were powdered and extracted with various solvent. The appearance and percent yield of extracts are reported as follows:

Table No. 3: Yield of various extract obtained from the flowers of *E.echinatus.*

Sr. No	Various Solvent used for Extraction	Colour	Nature	Percentage yield (% w/w)
1.	Methanol	Dark Greenish	Jelly like	7.5% w/w
2.	Chloroform	Redish Brown	Semisolid	4.5% w/w
3.	Aqueous	Dark Brown	Sticky Powder	9% w/w

Preliminary Phytochemical Screening of Various Extracts of *E. echinatus*

From the above test performed and observed show the Resut as follows

Test: + Indicates Presence of phytoconstituent - Indicates absence of phytoconstituents

Table 4: Phytoconstituents of Flower of *E.echinatus.*

Extracts	Methanol	Chloroform	Aqueous
Tests for carbohydrates			
Molish Test	+	+	+
Fehling Test	+	+	-
Benedict Test	-	+	-
Test for Monosaccharide			
Barfoed's Test	+	+	-
Test for non-reducing polysaccharides			
Iodine Test	-	-	-
Test for Proteins			
Biuret test	-	-	+
Millions test	-	-	-
Tests for Steroids			
Salkowaski reaction	+	-	-
Libermann Burchard reaction	+	-	-
Libermann reaction	+	-	-
Test for Glycosides			
Borntrager's Test	+	+	-
Killer- Killani Test	-	+	-
Test for Saponin			
Foam test	-	-	-
Tests for Flavonoids			
Shinoda test	+	-	-
Lead acetate Test	+	-	_
Sod-hydroxide Test	+	-	-
Tests for Alkaloids			
Meyers Test	+	-	-
Wagner's Test	-	+	-
Hager's Test	-	-	-
Dragendorff Test	+	+	-
Test for Tannins & Phenolic compounds			
$FeCl_3$	+	-	-
Lead acetate	+	-	-

Pharmacological Screening

1. Locomotar Activity: By using Actophotometer

Effect of various extract of flower of E.echinatuson mice by locomotors activity test as follows

Table No. 5: Locomotar Activity of mice.

Sr.no.	Treatment	Group of mice	Dose	Wt. of mice	Time spent	Before administration of drug	After 60min of administration of treatment
1.	Control	1	0.7mg/kg	14g	5min	86	86
2.	Std	2	0.75mg/kg	15g	5min	48	8*

	(Diazepam)						
3.	Methanol	3	0.85mg/kg	17g	5min	48	20*
4.	Chloroform	4	0.75mg/kg	15g	5min	160	127
5.	Aqueous	5	0.85mg/kg	17g	5min	58	23*

From the above observation we can conclude that methanolic extract and chloroform extract of *E. echinatus* show potent locomotor activity as compared to standard drug Diazepam.

2. **Antianxiety activity: By Using Elevated plus maze**

Effect of various extract of flower of *E.echinatuson* mice by Antianxiety activity test as follows

Table No. 6: Antianxiety Activity of Mice.

Sr.no.	Treatment	Group of mice	Dose Mg/kg	Wt. of mice	Time spent	Time spent in open Aarm (Sec.)	Time spent in closed Aarm (sec.)
1.	Control	1	0.7mg/kg	14g	5min	51	230
2.	Std (Diazepam)	2	0.75mg/kg	15g	5min	300*	0
3.	Methanol	3	0.85mg/kg	17g	5min	198	90
4.	Chloroforom	4	0.75mg/kg	15g	5min	300*	08
5.	Aqueous	5	0.85mg/kg	17g	5min	294*	12

From the above observation we can conclude that Animal from group 4 (Chloroform) spend more time in open Aarm as compared to group 3 and 5 Animal. From group 3, 4,5 spent more time in open Aram as compared to closed Aram hence the methanolic, chloroform & aqueous extract of drug E. echinatus have antianxiety activity. Chloroform extract group shows potent activity compared with standard.

3. Phenobarbitone induced sleeping time

Effect of various extracts of flower of *E. echinatus* mice by phenobarbitone induced sleeping time test as follows

Table No. 7: Recorded Sleeping Time of Mice.

Sr. no.	Body wt.	Treatment	Dose Mg/kg	Root Of admi.	Time of injection (Ti)	Time of sleep (Ts)	Time of wake (Tw)	Onset of action (Ts-Ti)	Duration of action
1.	14g	Phenobarbitone	0.7mg/kg	i. p.	11:30	12:45	2:00	75min	75min*
2.	15g	Pb.+ methanolic extract	0.75mg/kg	i. p.	11:45	12:55	1:55	65min	70min
3.	17g	Pb.+ Chloroform extract	0.85mg/kg	i. p.	11:50	1:12	2:42	72min	90min*
4.	15g	Pb. + Aqueous extract	0.75mg/kg	i. p.	12:00	12:56	2.4	56min	72min

From the above observation we can conclude that the methanol, chloroform and Aqueous extract of flower of *Echinopus echinatus* shows the Hypotonic Activity. Chloroform extract shows good result as compared with other extract and it potentiate Phenobarbitone induced sleeping time.

Antimicrobial Activity By Using Cup and Plate Method

Effect of various extract of flower of E.echinatus by Antimicrobial activity shows zone of Inhibition as follows

Table No. 8: Zone of Inhibition (mm).

Extract	**Concentration** ug/ml	**Micro-organism and Minimum inhibitory concentration in mm (MIC)**		
		E.coli	***S.aureus***	***B.subtilis***
Aqueous	10ug/ml	-	-	-
	20mg/ml	-	-	-
	30mg/ml	-	-	-
	40mg/ml	13mm	11mm	9mm
	50mg/ml	**17mm**	**15mm**	**13mm**
Chloroform	10mg/ml	-	-	-
	20mg/ml	-	10	12
	30mg/ml	-	-	13
	40mg/ml	7mm	9mm	10mm
	50mg/ml	**16mm***	**19mm***	**18mm***
Methanol	10mg/ml	-	-	-
	20mg/ml	-	-	-
	30mg/ml	-	-	-
	40mg/ml	12mm	**14mm**	9mm
	50mg/ml	12mm	**14mm**	8mm
Standard	100µg/ml	**25mm***	**30mm***	**27mm***

From the above observation we can conclude that chloroform show (19mm, 18mm) as compared to standard drug Norfloxacin (29mm).in the Microorganism.

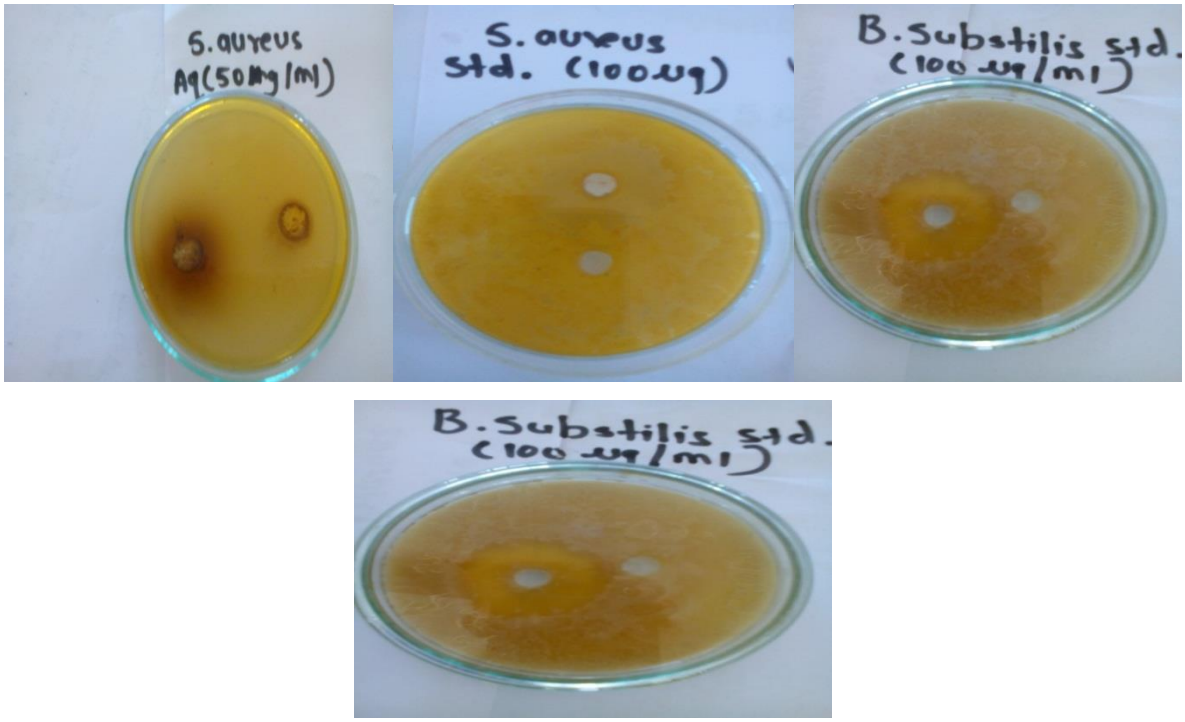

Fig. No. 5: All Fig Shows the Zone of Inhibition.

IV. CONCLUSION

The present study gives an idea about Phytochemical and Pharmacological Evaluation of flower of *Echinopus echinatus* Roxb.

- Preliminary Phytochemical study reveals that methanol Shows carbohydrate, steroid, flavonoid, glycoside and chloroform extract of flower shows the presence of the carbohydrates, steroid, Alkaloid, flavonoids, glycosides etc.
- Pharmacological evaluation of all extract for CNS depressant activity as compared to all extract chloroform extract shows good result in locomotors activity, Phenobarbitone induced Hypnotic Activity and Antianxiety activity. Chloroform shows significant result with compared to Diazepam and phenobarbitone as standard drug.
- Chloroform extract shows significant result with standard drug Norfloxacin.
- So from the above result chloroform extract of flower of *Echinopus echinatus* containing Phytochemicals like glycoside and alkaloid act as a **CNS depressant** and good **Antimicrobial activity.**
- In future it may be evaluated for various SAR study will result for good CNS Depressant Biomolecule.

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