Evaluation of CNS and Behavioral Activity of Evolvulus Nummularius Plant in Experimental Animal Models

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ABSTRACT

Aim: To evaluate Central Nervous System (CNS) and behavioral activity of EvolvulusNummularius plant in experimental animal models.
Methods: Experimental Animals (Rats) were acclimatized to laboratory conditions& were divided into four groups each containing 6 animals. Animals were divided into Control, standard, test-1, test-2 groups which receive 1% gum acacia, diazepam (10mg/kg), ethanolic extract of EvolvulusNummularius (200mg/kg) and (400mg/kg) respectively. Ethanolic extract of EvolvulusNummularius was used to examine CNS activity using experimental models like phenobarbitone sodium-induced sleeping time to evaluate sedative activity, Y-maze to evaluate exploratory behavior, Rotarod to evaluate muscle relaxant activity, and Actophotometer for locomotor activity.
Result & Discussion: Acute toxicity study revealed that no toxicity and mortality were seen and LD50> 2000mg/kg of extract. It is observed that no abnormality is seen during the assessment of the behavioral profile. Preliminary phytochemical analysis reveals the presence of chemical constituents like alkaloids, ergoline alkaloids, flavonoids, saponins, glycosides, anthraquinones, etc. which may responsible for psychopharmacological action. It is clear that plant extract significantly increases the sleeping latency and duration of sleep in rats when compared with control. It is observed that the extract showed a significant reduction in exploratory behavior of rats in y-maze when compared to control. In addition, the extract showed a marked reduction in muscle relaxant and locomotor activity of rats in Rotarod and Actophotometer respectively when compared with a control group.
Conclusion: Based on the result of the present study, it can be concluded that doses of 200mg/kg and 400mg/kg of ethanolic extract of aerial parts of EvolvulusNummularius possess CNS depressant activity.

Keywords: Central nervous system, Depressant, Extract, Sedative, Rotarod, Actophotometer, Y-maze, E. Nummularius.

INTRODUCTION

Herbal medicines are also called phytotherapy to refer to, using flowers, seeds, roots, stems, fruits, buds, and leaves for medicinal purposes. Herbalism has a long traditional use for centuries over conventional medicines. Herbal medicines consist of herbs and their various preparations, also herbal finished products containing active parts of a plant and other plant materials. These are mostly used for curing many diseases as well its prevention.[1] Despite medical and technological advancement, cultures around the world still relied upon traditional medicine to meet their healthcare needs. Around 75-80% of the population prefer and use traditional drugs because of a general belief that herbal drugs have minimal side effects and available at the lowest price. World health organization (WHO) reported that there is a two to three-fold increase in the use of herbal drugs.[2] Ayurveda, Siddha, Unani, Yoga, Naturopathy, and Homeopathy are the six recognized systems of medicine. It has evolved and continues serving the major population by providing effective, reliable, and affordable health services. To eradicate various side-effects of synthetic drugs, research has been developed findings of a drug through herbal resources. As it is expected that chemicals present in plants are safer and much cost-effective than the existing drug.[3]

Convolvulaceae, which is commonly known as bindweed or morning glory family, is a family comprising approximately 1,600–1,700 species grouped in 55–60 genera. The family is widely distributed in many countries, but its members are primarily tropical plants. Convolvulaceae plants are present in the form of herbaceous vines, but some plants are also in form of trees, shrubs, or herbs.[4]

EvolvulusNummularius of the Convolvulaceae family commonly known as round leaf bindweed is found in the northeastern state of India. It's a perennial herb with creeping stems and small rounded leaves alternatively arranged on the stems. Stems are prostrate, pubescent and nodes at rooting. Leaves are orbicular- obovate with 5-20mm long and broad with obtuse apex and truncate to subcordate base. Flowers are solitary or rarely paired, pedicels 2-2.5 mm long, up to 5 mm, and reflexed when fruiting. Corolla is white with deeply 5-6 lobed and 5-6 diameter.[5] It contains Alkaloids, Flavonoids, Carbohydrates, Glycosides, Tannins, Terpenoids, Cardinolides, Anthroquinones, Phenolic Compounds,[6]β-Sigmosterol, Stigmasterol, β-Sigmosterol Glucoside, Ursolic acid, Oleanolic acid, and d-Mannitol. It also contains 3-O (4-stearoyl-Zcoumaroyl)-stigmast5, E-22 dien-3ß-ol; 16-(Ecoumaroyloxy)-palmitic acid and 3ß-hydroxyurs-12-en-29ßoic acid.[7-8] It has medicinal uses like Antibacterial activity, Antioxidant activity, Anthelmintic, Fever, Wounds, Burns, Hysteria, Convulsion, Amoebic Dysentery, Anti-proliferative, and Paralysis.[8-12]In the 1960s, Hofmann and Tschetter stated that ergoline alkaloids are present in higher plants, especially in some Convolvulaceae plants. Since then, a large number of genera belonging to Convolvulaceae, including Argyreia, Ipomoea, and Stictocardia, was searched for the

presence of ergoline alkaloids, based on the idea that ergoline alkaloids are the main bioactive constituents responsible for the psychotomimetic effects of Convolvulaceae plants.(4) Further, no study regarding CNS activity has been performedSo the motive of the study is to evaluate the CNS and behavioral effects on EvolvulusNummularius in an experimental animal model to come up with the scientific pre-clinical establishment.

MATERIALS AND METHODS

Plant material and preparation of extract

Fresh plant of EvolvulusNummularius is collected from Saputara, the west Indian state of Gujarat, during November 2020. It was thoroughly cleaned and dried at room temperature in shade and kept away from direct sunlight. The plant was authenticated in the Department of biology, B.K.M. Science College, Valsad, Gujarat. The plant was collected in bulk and was washed with running tap water to remove soil followed by separation of aerial parts and was dried in shade. The dried plant materials werehomogenized into a fine powder and stored in an airtight container until use. The aerial part powder was successfully extracted by a soxhlet extractor using ethanol as solvent.And extraction was completed after 72 hours or when the extract becomes colorless. The extract was prepared using 1% gum acacia in normal saline[9] and administered to the animals in appropriate dose levels by the oral route of administration.

Animals

Wistar rats of either sex weighing 150-250 grams obtained from JAI research foundation Vapi Gujarat were used. All the rats were housed in the animal house at room temperature (22±30◦C) and humidity (30-70%). All the animals were treated as per the internationally accepted ethical guidelines for the care of laboratory animals. Before the experiments, rats were fed with standard food and distilled water ad libitum and were acclimatized to standard laboratory conditions and maintained at 12:12 hr light: dark cycle. All experiments and protocols described in the present study were approved by the Institutional Animal Ethics committee (IAEC) of ROFEL, Shri G.M. Bilakhia College of Pharmacy, and were performed as per CPCSEA norms.

Phytochemical characterization

The extracts were subjected to general phytochemical analysis for the presence of carbohydrates, proteins, amino acids, tannins, phenolics, flavonoids, alkaloids, ergoline alkaloids, anthraquinones, glycosides, saponin, and steroids using the standard methods.[13]

Drugs and Chemicals

Diazepam (Abbott Healthcare Pvt Ltd.), phenobarbitone sodium injection (NitinLifesciences Ltd), ethanol, gum acacia, and all other chemicals were of high quality and purity which was obtained from ROFEL, Vapi, Gujarat.

Acute toxicity studies (LD_{50})

The acute oral toxicity of EvolvulusNummularius was performed using The Organisation for Economic Cooperation and Development (OECD) 423 guideline- toxic class method.[14] The test substance was orally administered at dose 50,300,500,2000mg/kg body weight using three animals of single-sex. The animals will be observed for 14 days under the following profiles:

Undisturbed behavior: Body position, Sedation, Piloerection, Locomotor activity, Excitation, Ptosis, Exophthalmos.
Unusual Behavior: Writhing, Respiration, Tremors, Twitching, Convulsions, Startle.
With Handling: Body tone, Abdominal tone, Aggressive to the handler, Grip strength, Tail suspension, Corneal reflex, Lacrimation, Salivation, Urination, Defecation, Transfer arousal, Touch escape, Position struggle, Grasp irritability, Provoke biting.
In Open field: Ataxia, Tail elevation, Tail pinch, Finger withdrawal, Figure approach.

Animal grouping and treatment

For all the activities, Animals were divided into four groups except for the assessment of general behavior profile. Group I for Control, Group II for standard drug, Group III for Test-1 (low dose of ethanolic extract of EvolvulusNummularius, 200mg/kg), Group IV for Test -2 (high dose of ethanolic extract of EvolvulusNummularius, 400mg/kg).

General Behaviour Profile

Evaluation of general behavior profile was evaluated using the functional observation battery (FOB) test or Irwin test.[15-16] The parameters were evaluated during acute toxicological study at the interval of 30 minutes after drug administration followed by 1 hour after every 4hour.

UNDISTURBED BEHAVIOUR

For assessment of Body position, the cage was left undisturbed and the body position of the animals was recorded and scored. If the body is in the Completely flattened position it is scored 0, if Lying on one side then scored 1, Lying upright = 2, Sitting crouched or sitting up = 3, Standing upright on hind limb = 4, Repeated vertical jumping = 5

For assessment of Locomotor activity,Rats were placed in an open field and were observed for 1 min in terms of their speed and movement and were scored accordingly. If rats are None resting then it scored 0, if rats Casual Scratch & Groom/slow spatial then scored 1, Vigorous Scratch & Groom/moderate spatial = 2, Vigorous movement = 3, Extremely vigorous movement = 4

UNUSUAL BEHAVIOUR

For assessment of Writhing, abdominal stretching of animals was observed. If no writhing is seen it scored as 0, Slight = 1 Moderate = 2 Marked = 3 Extreme = 4

For assessment ofthe Startle response sudden sound was made, following a sharp noise an animal should startle by immediate freezing. If no startle then it is scored as 0, if Slight response =1, Normal = 2, Exaggerated response =3

WITH HANDLING

For assessment of Touch Escape,the body of the animal was touched from the region of the lower thorax at least thrice. The escape response was scored as the speed of animal as below: if Slow or slight escape scored 0, Moderately escape = 1, Vigorously rapid escape = 2, Extreme vigorous running = 3

For assessment of Grip strength,An animal was held gently by the tail. Place on wired mesh (holding the tail) & is pulled slowly horizontally, backward (1 sec.) and was scored as if no resistance to pull then scored 0, if Slight then scored 1, Moderate = 2, Active = 3, Unusually effective = 4

IN OPEN FIELD

In a tail pinch, animals were placed in an open field and pressure was applied on the tail for 4sec and was scored accordingly.if No response then scored 0, A very slight movement, slight freezing = 1, Slight biting escape/moderate freeze = 2, Moderate bite escape/ abrupt active freeze = 3, Vigorous biting escape = 4, Extreme Vigorous biting escape = 5

Assessment of Phenobarbitone sodium-induced sleeping time

Healthy Wistar rats each weighing 150-200grams are used to evaluate sedative activity. Experimental rats were divided into three groups each containing six animals. All the healthy Wistar rats fasted for 24 hours before experiments. Then for assessment of activity GROUP I received vehicle (1% gum acacia in normal saline), GROUP II received 200mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius, GROUP III received 400mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius. After 30 minutes of test drug administration, all the groups received phenobarbitone sodium injection (50mg/kg) intraperitoneally. Animals were placed back to their cages and the duration of loss of righting reflexes and duration of sleep was measured as the time interval between loss and regain of righting reflux.[17]

Assessment of exploratory behavior using Y-MAZE

In y-maze, behavioral assessment of rodents is carried out by measuring the willingness of rodents to explore new environments. Usually, rodents prefer to investigate new arms rather than explored previously.

Healthy Wistar rats each weighing 150-200grams were divided into four groups each containing six animals. Y-maze is a Y-shaped maze with three arms which is white opaque aiming at a 120o angle from each other. In these tests, initially, rats were explored to all three arms as a trial then an actual test was performed in each group of 6 Wistar rats at 30, 60, 90, and 120 min after treatment of GROUP I which received vehicle (1% gum acacia in normal saline), GROUP II received 10mg/kg Diazepam (p.o.), GROUP III received 200mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius, GROUP IV received 400mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius. Then rats were again placed individually in a symmetrical Y-shaped runway (33 × 38 × 13cm) for 3 min and the number of times a rat entered in the arm of the maze with all 4ft (an 'entry') were counted.[17]

Assessment of muscle relaxant activity using ROTAROD

Rotarod consists of a horizontally oriented revolving rod which is mounted above the floor which is low enough to avoid injury and at a particular height to avoid easily fall of rodents. Rods are 3cm in diameter which revolves at 25 rpm. The principle of rotarod is to evaluate the effects of drugs acting on motor coordination, balance, and motor learning in rodents. The skeletal muscle relaxation induced by test compounds can be evaluated by testing the ability of a rat to remain on a revolving rod

Healthy Wistar rats each weighing 150-200grams were divided into four groups each containing six animals In this test, rats will be initially placed on the rotarod and the rats that maintain balance for (3 sec) in three successive trials were included in the experiment. After the trial, GROUP I received vehicle (1% gum acacia in normal saline), GROUP II received 10mg/kg Diazepam (p.o.), GROUP III received 200mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius, GROUP IV: received 400mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius. After 30 minutes of oral administration, the rats were placed on the rod followed by 60,90, and 120 minutes. The fall-off time of rats was recorded and then the percentage of the animal falling from the rotarod within the test period for every concentration was calculated.[17]

Assessment of locomotor activity using ACTOPHOTOMETER

It consists of a hollow square chamber and grills of nickel-plated brass rods at the bottom which are placed 3/16" apart. It also contains the counter chamber in which the movement of animals can be counted with the help of six photocell beams. The locomotor activity can be analyzed when a beam of light is cut by rodents.

Healthy Wistar rats each weighing 150-200grams were divided into four groups each containing six animals In this experiment, every rat was observed for 5 min in a square closed field area (30 cm × 30 cm × 30 cm) equipped with six photocells in the outer wall. Interruptions of photocell beams (locomotor activity) were recorded. The actophotometer was turned on and each rat was placed individually in the activity cage for 5 min to check the locomotor activity. The basal activity score for all the animals was noted. After then GROUP I received vehicle (1% gum acacia in normal saline), GROUP II received 10mg/kg Diazepam (p.o.), GROUP III received 200mg/kg, (p.o.) ethanolic extract of EvolvulusNummularius, GROUP IV received 400mg/kg, (p.o.) ethanolic

extract of EvolvulusNummularius. Then after 30, 60, 90, and 120 minutes, the activity score for 5 min was again observed. The difference in the activity, before and after drug administration, was noted down. [17]

STATISTICAL ANALYSIS

The data of all the parameters were analyzed for differences among the control/treated group. The result of the study was expressed as mean ± SEM and was analyzed by one-way ANOVA with pair t-test, and $p < 0.05$ was considered statistically significant

RESULTS AND DISCUSSION

Phytochemical analysis

The percentage yield of EvolvulusNummularius was 9.6%w/w. The preliminary phytochemical screening of the EvolvulusNummularius extractshowed the presence of alkaloids, ergoline alkaloids, carbohydrates, flavonoids, saponins, protein, and anthraquinones. All the extracts were placed in an airtight container for further pharmacological studies.

Acute toxicity study

Acute oral toxicity study of ethanolic extract of EvolvulusNummularius was carried out as per OECD 423 guidelines i.e. Toxic class method. The study revealed that no toxicity and mortality were seen and LD50> 2000mg/kg of extract.

General behavior profile

The result obtained through the functional observational battery test is present in Table 1. The extracts affected the locomotor activity as well as grip strength. Slight sedation was also observed with weak transfer arousal. The results indicate that the extract influences general behavioral profiles, as evidence in the spontaneous activity, touch, and pain responses.

Table: 1 Behavioural profile of rats

Sr.No.	Parameter	Score
A	**UNDISTURBED BEHAVIOUR**	
1	Body position	**3** (sitting up)
2	Locomotor activity	**1** (Casual Scratch & Groom/slow spatial)
3	Sedation	Yes
4	Excitation	No
5	Pilo erection	**0** (normal)
6	Ptosis	**0** (normal)
B	**UNUSUAL BEHAVIOUR**	
1	Respiration	**0** (normal)
2	Tremors	**0** (none)
3	Convulsion	**0** (none)
4	Writhing	**0** (none)
5	Twitching	**0** (normal)
C	**STEREOTYPE CHANGES WITH HANDLING**	
1	Aggressive to the handler	**0** (normal)
2	Grip strength	**1** (slight)
3	Corneal reflex	**0** (none)
4	Lacrimation	**No**
5	Salivation	**0** (none)
6	Urination and Defecation	**Yes**
7	Transfer Arousal	**2** (Moderate dull, relaxed appearance, moderate slow movement)
8	Touch escape	**0** (slight escape)

Hypnotic and sedative activity

The sedative activity of ethanolic extract of EvolvulusNummularius was evaluated using phenobarbitone-sodium-induced sleeping time. Two parameters that were evaluated are sleep latency and duration of sleep. The results (table-2) showed that sleep latency produced by control, Test-1(200mg/kg) and Test-2 (400mg/kg) are 7.33±0.91, 6.83±0.74 and 4.33±0.55$^{*\#}$ minutes respectively. Similarly, duration of sleep (minutes) is prolonged in Test-1 and Test-2 groups 39.66±1.83**, 54.33±1.4$^{**\#}$ respectively when compared to control 12.33±1.4. So it can be said that Test groups have potentiated the phenobarbitone sodium-induced sleeping time as well as the duration of sleep increases in a dose-dependent manner in an experimental animal model. The sedation produces by EvolvulusNummularius in phenobarbitone-induced sleeping time it might be due to its CNS depression or tranquilizing action.

Table: 2 Effect of EvolvulusNummularius extract on the sedative activity of rats

Groups	Dose	Sleep latency (minutes)	Duration of sleep (minutes)
Control	1% gum acacia + phenobarbitone (i.p.)	7.33±0.91	12.33±0.91
Test-1	200mg/kg E. Nummularius + phenobarbitone (i.p.)	6.83±0.74	39.66±1.83**
Test-2	400mg/kg E. Nummularius + phenobarbitone (i.p.)	4.33±0.55$^{*\#}$	54.33±1.4$^{**\#}$

All values are expressed as mean ± S.E.M.; (n=6) animals in each group
Data were analyzed by one-way ANOVA followed by paired t-test.
*P<0.05 and**P<0.01 when compared with control.
$^{\#}$P<0.01 when compared with Test-1

GRAPHICAL REPRESENTATION

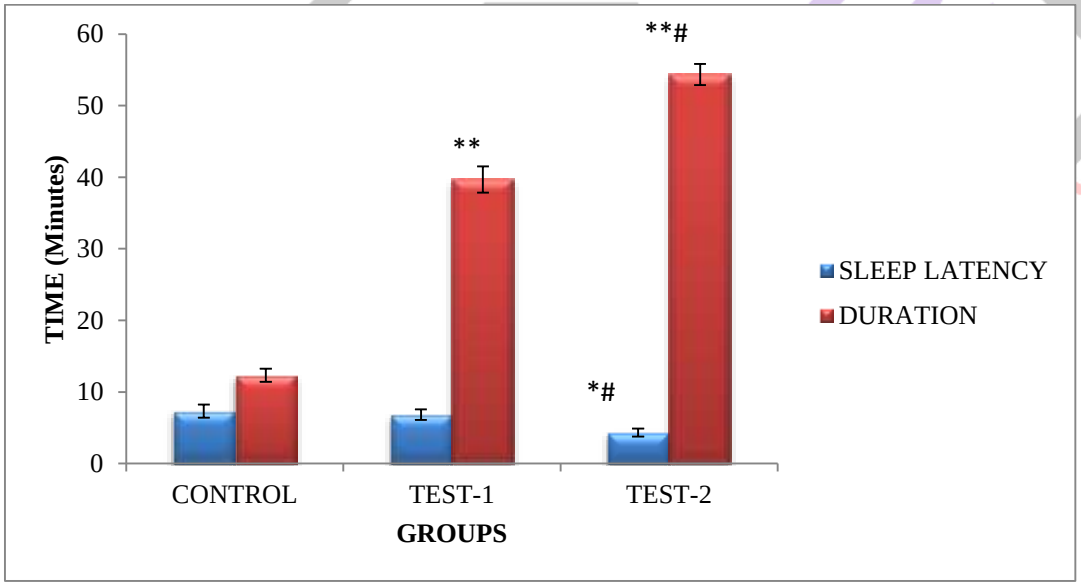

Figure: 1 Graphical representation of effects of E. Nummularius extract on the sedative activity

Exploratory behavior

Exploratory behavior of ethanolic extract of EvolvulusNummularius was evaluated using Y-maze. The number of entries in the new environment is seen to evaluate exploratory behavior. Results (Table 3) showed that there is a decrease in the number of entries of the standard group when compared with the control. It is also seen that as time increases the effect of E. Nummularius increases i.e. the number of entries in the new environment decreases.

Both the doses 200mg/kg and 400mg/kg showed a significant decrease in the visit of arms of Y-maze when compared with control and standard. Exploratory behavior of extract of E. Nummularius in an experimental animal model is in a dose-dependent manner.

Table: 3 Effect of EvolvulusNummularius extract on exploratory behavior of rats

Groups	Dose	Exploratory behavior observed for 5 minutes (Number of entries in new environment)			
		30	60	90	120
Control	1% gum acacia	11.1±0.3	9.83±0.16	9.66±0.2	8.66±0.21
Standard	Diazepam (10mg/kg)	7.6±0.5**	7.3±0.5**	6.5±0.2**	1.6±0.21**
Test-1	200mg/kg E. Nummularius	10.5±0.22#	10±0.4#	8±0.4*#	6±0.22**#
Test-2	400mg/kg E. Nummularius	10.3±0.5#	9±0.63	6±0.36**$	2.3±0.21**$

All values are expressed as mean ± S.E.M.; (n=6) animals in each group
Data were analyzed by one-way ANOVA followed by paired t-test.
*P<0.05 and **P<0.01 when compared with control.
#P<0.01 when compared with standard
$P<0.01 when compared with Test-1

GRAPHICAL REPRESENTATION

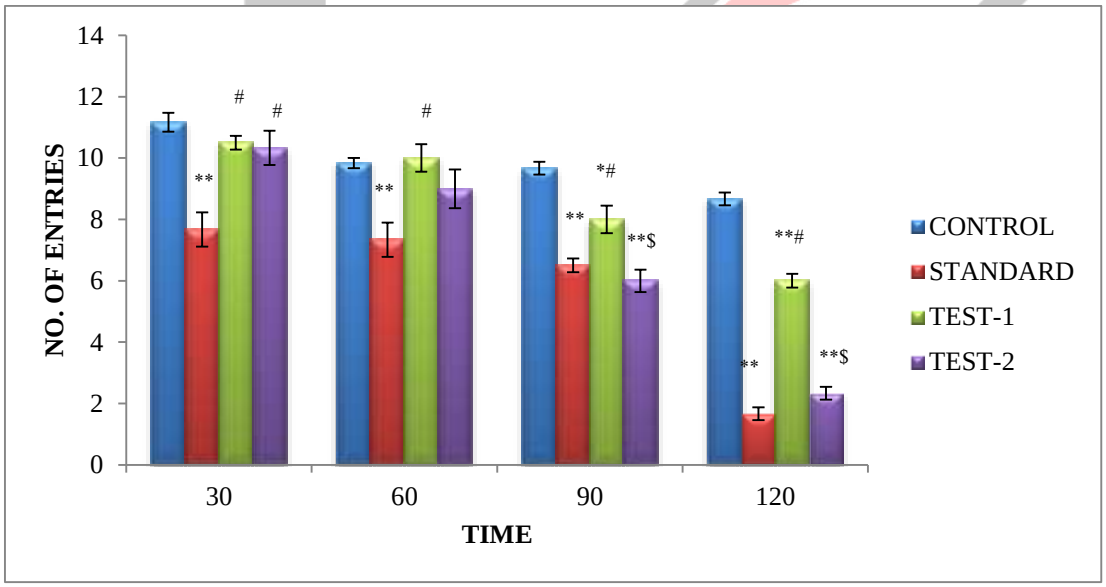

Figure: 2 Graphical representation of effects of E. Nummularius extract on exploratory behavior.

Muscle Relaxant Activity

The myorelaxant activity of EvolvulusNummularius was evaluated using the Rotarod method. The fall of time parameter was evaluated and was analyzed at different time intervalsfor 5 minutes. Results (Table 4) showed that falling of time increases in standard and test group when compared to control group after administration of a drug.

It's seen that test groups (200mg/kg and 400mg/kg) significantly reduced the time spent on the revolving rod when compared with control (<0.01). Standard drug (Diazepam) also showed a significant reduction in fall of time when compared with control (<0.01).

We can say that the extract of E. Nummularius produced a significant reduction in motor coordination of tested rats in a dose-dependent manner.

Table: 4 Effect of EvolvulusNummularius extract on muscle relaxant activity of rats

Groups	Dose	Muscle relaxant activity observed for 5 minutes (fall of time in seconds)			
		30	60	90	120
Control	1% gum acacia	282.5±1.11	277.16±1.3	252.5±1.7	233.3±1.6
Standard	Diazepam (10mg/kg)	145±1.09**	137.6±0.91**	86.83±0.7**	53.33±0.55**
Test-1	200mg/kg E. Nummularius	272.5±0.92**#	252±0.96**#	198±0.96**#	151.1±0.54**#
Test-2	400mg/kg E. Nummularius	152.5±0.9**#$	140±0.68**#$	92.6±0.76**#$	61.3±0.33**#$

All values are expressed as mean ± S.E.M.; (n=6) animals in each group
Data were analyzed by one-way ANOVA followed by paired t-test.
*P<0.05 and **P<0.01 when compared with control.
#P<0.01 when compared with standard
$P<0.01 when compared with Test-1

GRAPHICAL REPRESENTATION

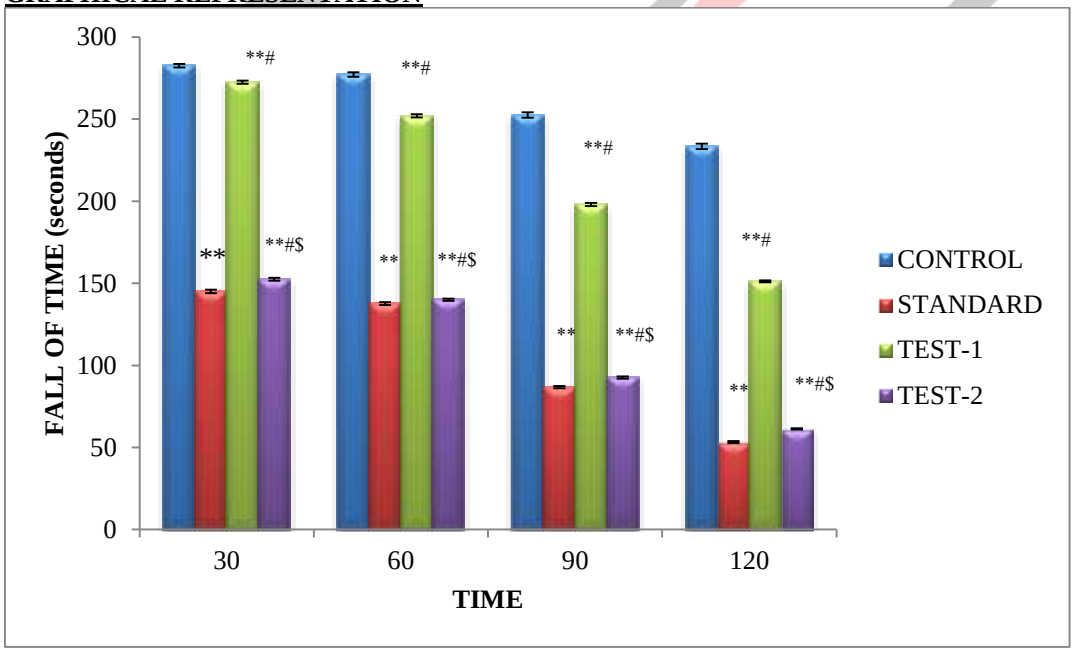

Figure: 3 Graphical representation of effects of E. Nummularius extract on muscle relaxant activity

Locomotor activity

The locomotor activity of EvolvulusNummularius was evaluated using the Actophotometer method. Cut-off time is the evaluation parameter. After drug administration locomotor activity decreases in Test-1 62.33±0.9 to 50±1.8, Test-2 60.67±0.4 to 45±1.8, standard 61.33±0.4 to 32.6±1.1 and it goes on reduction as time for observation increases up to 120 minutes. Results (Table 5) showed that Test-1 (200mg/kg) shows moderate reduction while Test-2 (400mg/kg) shows significant reduction when compared with control (<0.01). E. Nummularius extract showed locomotor activity in a dose-dependent manner.

Table: 5 Effect of EvolvulusNummularius extract on locomotor activity of rats

Groups	Dose	Locomotor activity observed for 5 minutes (cut off time in seconds)				
		0	30	60	90	120
Control	1% gum acacia	61.83±0.8	59.33±0.4	57.5±1.12	51.83±0.7	49.67±0.33
Standard	Diazepam (10mg/kg)	61.33±0.4	32.6±1.1**	21±0.63**	17±0.97**	14.6±1.52**
Test-1	200mg/kg E. Nummularius	62.33±0.9	50±1.8**#	31.8±1.5**#	13.5±1.02**	8±0.73**#
Test-2	400mg/kg E. Nummularius	60.67±0.4	45±1.8**#	26.6±1.5**#	15±1.0**	6.67±0.4**#$

All values are expressed as mean ± S.E.M.; (n=6) animals in each group
Data were analyzed by one-way ANOVA followed by paired t-test.
*P<0.05and **P<0.01 when compared with control.
#P<0.01 when compared with standard
$P<0.01 when compared with Test-1

GRAPHICAL REPRESENTATION

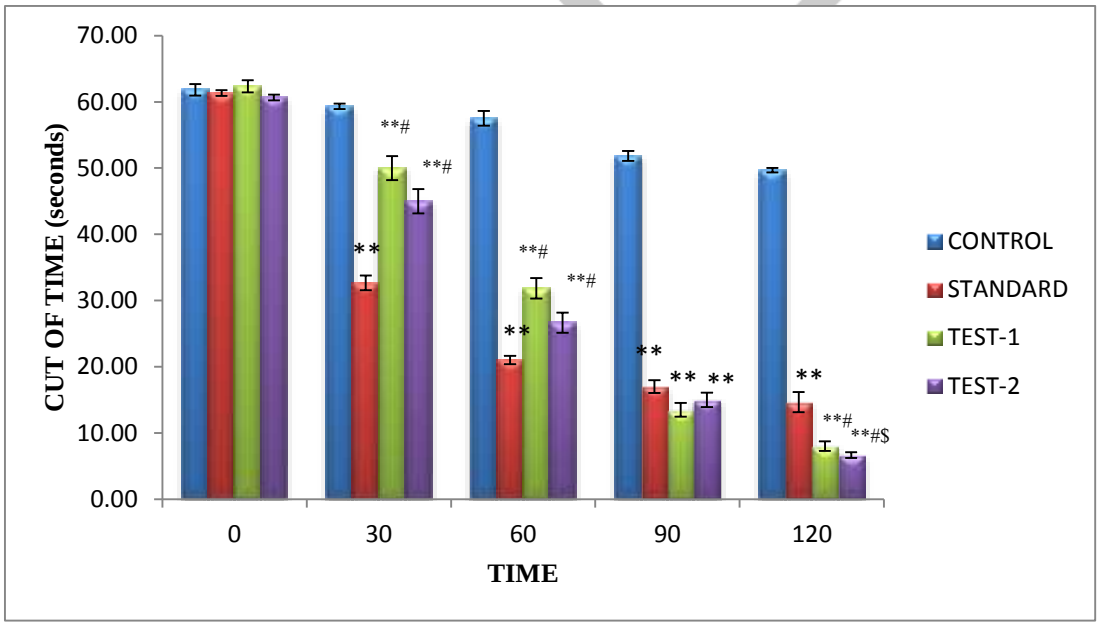

Figure: 4 Graphical representation of effects of E. Nummularius extract on locomotor activity

CONCLUSION

EvolvulusNummularius is a perennial herb that belongs to the family Convolvulaceae which was used to examine preliminary phytochemical and psychopharmacological action. The screening of phytochemical showed the presence of chemical constituents like alkaloids, ergoline alkaloids, glycosides, saponins, tannins, anthraquinones which may responsible for psychopharmacological action. Based on the result of the present study, it can be concluded that doses of 200mg/kg and 400mg/kg of ethanolic extract of aerial parts of EvolvulusNummularius possess CNS depressant activity. The extract showed potentiation of sleep and duration of sleeping time. It also showed a marked reduction in locomotor, muscle relaxant activity and a significant decrease in exploratory activity. This might be due to various mechanisms acting on receptors including glycine and brain GABA receptors. The ability of the extract to potentiate the sedative activity may work by interacting with GABA. Hence the ethanolic extract of EvolvulusNummularius shows CNS depressant action either by directly activating the GABA receptor or by enhancing GABA action. However, further studies are required to know the exact mechanism of CNS depressant effects and to examine the active constituent responsible for showing these pharmacological activities.

REFERENCES

[1] Sahoo N, Manchikanti P, et al. "Herbal drugs: standards and regulation". *Fitoterapia*. **2010**; 81(6), 462-471.

[2] Gunjan M, Naing TW, et al. "Marketing trends & future prospects of herbal medicine in the treatment of various disease". *World J of Pharmaceutical Research.***2015**; 4(9), 132-155.

[3] Ravishankar B, Shukla V, et al. "Indian systems of medicine: a brief profile". *Afr J Tradit Complement Altern Med*. **2007**; 4(3), 319-337.

[4] Chen GT, Lu Y, et.al., "Medicinal uses, pharmacology, and phytochemistry of Convolvulaceae plants with central nervous system efficacies: A systematic review". *Phytotherapy Research*. **2018** May; 32(5), 823-64.

[5] Chen S-H, Su J-Y, et al. "Notes on two newly naturalized plants in Taiwan: Evolvulusnummularius (L.) L.(Convolvulaceae) and AcalyphaaristataKunth (Euphorbiaceae)". *J Taiwania*. **2009**; 54(3), 273-278.

[6] Tamilarasan M, Sivamani S,et.al.,"Phytochemical Screening and In Vitro Bioactivities of the Extracts of Aerial Part of Evolvulusnummularius". *Intern. J.ofpharmaco.& phytochemical Res.***2015**;7(1);111-116

[7] Sahu PK, Gupta SJB. "Medicinal plants of morning glory: convolvulaceaeJuss. of central India (Madhya Pradesh and Chhattishgarh)". **2014**; 2(2), 463- 469.

[8] Dinda B, Ghosh B, et.al.,"Chemical constituents of Evolvulus nummularius" *Indian J. of Chemistry*.**2007,** 46B(03), 492-498

[9] Mandarm, karkasr, et.al.,"Evaluation of anti-pyretic and woundhealing activities of evolvulus nummularius leaves in albino wistar rats".*Int j curr pharm res*.**2015**; 7(1), 74-5.

[10] Pavithra PS, Sreevidya N, et.al.,"Antibacterial and antioxidant activity of methanol extract of Evolvulus nummularius". *Indian J. of pharmacol*. **2009** Oct;41(5), 233.

[11] Rupanjali S, Biswanath D, et.al.,"Anti-proliferative activity of Evolvulus nummularius, a medicinal plant of Tripura, on doxorubicin resistant leukemia cells."*Journal of Applied Bioscience*. **2010;** 36(2), 161-3.

[12] Iqbal I, Shabbir A, et al. "Evolvulus nummularius (L.) L.(Convolvulaceae): a new alien plant record for Pakistan". **2020**; 9(4), 702-711.

[13] Khandelwal K. Practical pharmacognosy. Pragati Books Pvt. Ltd.; 2008 Sep 7.

[14] Kushwaha S, Dashora A, et al. "Acute oral toxicity studies of the standardized methanolic extract of PhyllanthusamarusSchum&Thonn". J *of pharma research.***2013**; 6(7), 720-724.

[15] Mathiasen J. R., & Moser V.C., "The Irwin Test and Functional Observational Battery (FOB) for Assessing the Effect of Compounds on behaviour, physiology, and safety Pharmacology in Rodents", *Current Protocols in Pharmacology,Wiley Online Library*,**2018;** 83(1), 1-18.

[16] Moser VC, "Neurobehavioral Screening in Rodents", *Current Protocol in Toxicology*, **1999**, 11.2.1- 11.2.16.

[17] C.A.Sureshkumar, et.al., "Psychopharmacological studies on the stem of Saccharumspontaneum", *International Journal of PharmTech Research***2010,** Jan-Mar, 319-321