Original Research Article

Evaluation of Adaptogenic and Antidepressant Activity of Rhododendron arboreum flower

ABSTRACT

Several medicine are available for the treatment of stress & depression, drugs of plant origin are gaining popularity due to their effectiveness with minimum side effect. Hence evaluation of adaptogenic and antidepressant effect of *Rhododendrom arboreum* flowers was carried out in different animal models. The adaptogenic activity was studied in anoxia stress, swimming endurance stress induced models in mice and rats. In anoxia stress animals were sealed in hermetic vessels and the increased in anoxia tolerance time was considered as positive effects. In swimming endurance model various biochemical parameters like blood glucose, cholesterol, triglycerides and Blood Urea Nitrogen, blood cell count like Red Blood Cells, White Blood Cells, and Differential Leukocyte Count were estimated; weight of organs like liver spleen and adrenal gland was measured in stress control and extracted treated animals. Reduction in biochemical parameters by the extracted shows the adaptogenic activity. Protection in deviation of blood cell counts by the extracts the adaptogenic activity. Protection effect against increased weight of liver, adrenal gland and decreased weight of spleen in stress conditions has revealed the adaptogenic activity of ethanolic extracts of *Rhododendrom arboreum* flowers. Immobility as behavioral parameter was assessed in tail suspension test and elevated plus maze model of depression after treatments with ethanolic extract of *Rhododendrom arboreum* flower at selected dose of 200mg/kg and 400mg/kg. The results have shown significant adaptogenic and antidepressant activity of Rhododendrom *arboreum.* Ethanolic extract of *Rhododendrom arboreum at* higher dose has shown significant activity than lower dose.

Keywords: Rhododendrom arboreum; adaptogenic activity; anoxia stress tolerance; antidepressant activity; tail suspension test.

1. INTRODUCTION

Stress is a common phenomenon that is experienced by every individual. When stress becomes extreme, it is harmful for the body and hence needs to be treated. Stress is involved in the pathogenesis of a variety of diseases that includes psychiatric disorders such as depression and anxiety immuno-suppression, endocrine disorders including diabetes mellitus, male impotence, cognitive dysfunction, peptic ulcer, hypertension and ulcerative colitis [1].

Stress is basically a reaction of mind and body against change in the homeostasis. The productive stress is called Eustress while harmful stress is called Distress. If the stress is extreme, the homeostatic mechanisms of the organism become deficit and the survival of the organism is threatened. Stress alters the equilibrium of various hormones which have a significant impact on the immune response in general. The status of immune system immuno-suppression versus immune-potentiation will depend upon the net effect of these changes [2].

1.1 General adaptation syndrome

Physiologists define stress as how the body reacts to a stressor, real or imagined a stimulus that causes stress. Acute stressors affect an organism in the short term; chronic stressors over the longer term. Selye researched the effects of stress.

Alarm is the first stage. When the threat or stressor is identified or realized, the body's stress response is a state of alarm. During this stage adrenaline will be produced in order to bring about the fight-or-flight response. There is also some activation of the HPA axis, producing cortisol.

Resistance is the second stage. If the stressor persists, it becomes necessary to attempt some means of coping with the stress. Although the body begins to try to adapt to the strains or demands of the environment, the body cannot keep this up indefinitely, so its resources are gradually depleted.

Exhaustion is the third and final stage in the GAS model. At this point, all of the body's resources are eventually depleted and the body is unable to maintain normal function. The initial autonomic nervous system symptoms may reappear (sweating, raised heart rate etc.). If stage three is extended, long term damage may result as the body, and the immune system is exhausted and function is impaired resulting in decompensation. The result can manifest itself in obvious illnesses such as ulcers, depression, diabetes, trouble with the digestive system or even cardiovascular problems, along with other mental illnesses [3].

Depression is a significant contributor to the global burden of disease and affects people in all communities across the world. Today, depression is estimated to affect 350 million people. The World Mental Health Survey conducted in 17 countries found that on average about 1 in 20 people reported having an episode of depression in the previous year. Depressive disorders often start at a young age; they reduce people's functioning and often are recurring. For these reasons, depression is the leading cause of disability worldwide in terms of total years lost due to disability. The demand for curbing depression and other mental health conditions is on the rise globally [4]. Depression is a common psychiatric disorder, with diverse symptoms and high co morbidity with other brain dysfunctions. Due to this complexity, little is known about the neural and genetic mechanisms involved in depression pathogenesis. In a large proportion of patients, current antidepressant treatments are often ineffective and /or have undesirable side effects, fueling the search for more effective drugs [5].
Depressive episode involves symptoms such as depressed mood, loss of interest and enjoyment, and increased fatigability. An individual with a mild depressive episode will have some difficulty in continuing with ordinary work and social activities, but will probably not cease to function completely. It is very unlikely that the sufferer will be able to continue with social work, or domestic activities, except to a very limited extent. Bipolar affective disorder typically consists of both manic and depressive episodes separated by periods of normal mood. Manic episodes involve elevated mood and increased energy, resulting in over-activity, pressure of speech and decreased need for sleep [6].

2. MATERIAL AND METHOD

2.1 Drugs and chemical used

Withania somnifera was procured from the Sapthagiri Pharma, Bangalore, India. *Imipramine* was procured from HCG pharma, Bangalore, India. All other reagents used were of analytical grade.

2.2 Experimental animals

Albino Wistar rats weighing between 160-220g gm and Albino mice weighing 20-30 gm of either sex were used for this purpose . The animals were randomized into experimental, normal and control groups, housed individually in sanitized polypropylene cages containing sterile paddy husk as bedding. The experimental animals were maintained under 12:12 h light dark cycle, in an animal house with controlled temperature (20-25ºC) and humidity. The animals were maintained under standard condition in an animal house approved by the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

2.3 Collection and authentication of plant material

The flowers of *R. arboretum* were collected from the higher altitude hilly region of Nepal. The plant was identified and authenticated by Dr. K. Ravi kumar, senior botanist at FRLHT (Foundation for Revitalisation of Local Health Traditions) JarakabandeKaval post,Attur, Yehalanka, Bangaluru (560106). A herbarium specimen was preserved in the college museum for future reference. The flower were shade dried at room temperature for 15 days and pulverized

2.4 Extraction procedure

The flowers of *R. arboretum* was dried under shade and then powdered with a mechanical grinder. The coarse powder of *R. arboretum* (500 gm) was subjected to extraction with 70% ethanol (1000 ml) after defatting with petroleum ether by Soxhelet extraction for the duration of 72 hours. During the process of extraction, the alternate filling and emptying of the body of the extractor goes on continuously till it gets exhausted and it was confirmed by discoloration of solvent in the side tube of extractor (Siphon). Then after, the residue was removed by filtration and concentrated under reduced pressure. The concentrated extract was transferred to china dishes and finally placed in a vacuum oven at 45°C. The extract was semisolid, brownish black in color and it was stored in an airtight amber colored bottle until used [7].

2.5 Preliminary Phytochemical Analysis

Ethanolic extract of thel flower of *R.arboreum* was subjected to chemical tests for the identification of their active constituents. Test for the presence of carbohydrates, glycosides, resins, tannin, alkaloid, fixed oil, flavonoids, terpenoids, protein, saponins, anthraquinone and amino acid were conducted as per the standard procedure [8].

2.6 Acute toxicity and dose selection [9, 10]

The maximum lethal dose of *R.arboreum* having the same chemical constituents was found to be 2000 mg/kg body weight, hence $1/10^{th}$ and $1/20^{th}$ of maximum lethal dose was taken as effective dose for the ethanolic extract of *R. arboreum* for adaptogenic and antidepressant activity

2.7 Pharmacological Screening

2.7.1 Evaluation of Adaptogenic activity of *Rhododendron arboreum* by anoxia stress tolerance test [11]

Albino mice of either sex weighing between 18-22 g are divided into four groups of six in each. Hermetic vessel of 500 ml air capacity is used for this test. Each animal is kept in the hermetic vessel and the time to show the first sign of convulsion is noted, it is immediately removed from the vessel and resuscitated if needed. After one week of drug treatment the animals are once again exposed to the anoxia stress.

Similarly the animals are also observed at the end of 2nd and 3rd weeks with the same treatment and the time duration for anoxia stress tolerance is noted

Experimental protocol

Animals were divided into four groups of six animals each. The groups were as follows:

Group 1:Normal control (mice administered with(,normal saline 10ml/kg b.w.p.o.) daily for 21 days.

Group 2: Standard *Withania somnifera* (animals were administered with single dose of *Withania somnifera* (100 mg/kg, p.o.) daily for 21 days.

Group 3: animals received single dose ethanolic extract of *R. arboreum (*200 mg/kg p.o. *)* daily for 21 days.

Group 4: animals received single dose of ethanolic extract of *R. arboreum (*400 mg/kg p.o. *)* daily for 21 days.

2.7.2 Evaluation of Adaptogenic activity of *Rhododendron arboreum* **by Swimming Endurance Test in Rats** [11].

Rats of either sex (150-200 g) are divided into 4 groups of six in each are used for the study. Stress is exerted in rats by keeping them in cylindrical vessels (length 48 cm and width 30 cm) filled with water to a height of 25 cm and the total swimming time for individual rat is noted, the rats are allowed to swim daily till exhausted. Extracts are given to rats once daily for a period of 14 days and on 15th day mean swimming time for each group is calculated, blood was collected through retro orbital plexus under light ether anesthesia to estimate biochemical parameters like blood glucose, triglycerides, cholesterol, BUN and blood cell count (RBC, WBC, DLC). Animals were sacrificed by cervical dislocation and the weight of organs such as liver, adrenals, spleen were recorded after washing with alcohol

Experimental protocol

Animals were divided into four groups of six animals each. The groups were as follows

Group 1: Normal control (rats administered with,normal saline 10ml/kg b.w.p.o.) daily for 14 days.

Group 2: Standard *Withania somnifera* (animals were administered with single dose of *Withania somnifera* (100 mg/kg, p.o.) daily for 14 days.

Group 3: Rats received single dose ethanolic extract of *R. arboreum (*200 mg/kg p.o. *)* daily for 14 days

Group 4: Animals received single dose of ethanolic extract of *R. arboreum (*400 mg/kg p.o. *)* daily for 14 days**.**

2.7.3 Evaluation of antidepressant activity of *Rhododendron arboreum* **by tail suspension test (TST) [12]**

Albino mice weighing about 20-30g are used. Animals were moved from their housing colony to laboratory in their own cages and allowed to adapt to the laboratory conditions for 1-2 hrs. Each animal was individually suspended in the wooden box with the help of hanging clip the animal was suspended 50 cm above the floor of that wooden box and the 1cm part of the tail was clipped. Each animal under test was both acoustically and visually isolated from other animals during test. The total period of immobility was recorded manually for 6 min. Each animal showed vigorous movement during initial 2 min period of the test. The duration of immobility was manually recorded during the next 4 min of the total 6 min testing period. Animal was considered to be immobile when it didn't show any body movement, hung passively and completely motionless. The test was conducted in a dim lighted room and each mouse was used only once in the test. After successive 14 days of treatment with control, standard and extract drugs, the immobility is calculated and recorded.

Experimental protocol

Animals were divided into four groups of six animals each. The groups were as follows:
Group 1:Normal control (mice administered with(,normal saline 10ml/kg b.w.p.o.) daily for 14 days
Group 2: Standard *Imipramine* (animals were administered with single dose of *Imipranime* (10 mg/kg, p.o.) daily for 14 days
Group 3: animals received single dose ethanolic extract of *R. arboreum (*200 mg/kg p.o. *)* daily for 14 days
Group 4: animals received single dose of ethanolic extract of *R. arboreum (*400 mg/kg p.o. *)* daily for 14 days

2.7.4 Evaluation of antidepressant activity of *Rhododendron arboreum* **by elevated Plus-Maze [13]**
The elevated plus-maze comprised two open (30 cm×5 cm×0.25 cm) and two enclosed (30 cm×5 cm×15 cm) arms that radiated from a central platform (5 cm×5 cm) to form a plus sign. The maze is constructed of black painted wood. A slight raised edge on the open arms (0.25 cm) provided additional grip for the animals. The plus-maze is elevated to a height of 40 cm above floor level by a single central support. The experiment is conducted during the dark phase of the light cycle (9:00–14:00 h). The trial is started by placing an animal on the central platform of the maze facing an open arm. The number of entries into, and the time spent in, each of the two types of arm, are counted during a 5 min test period. The percentage open arm entries and percentage open arm time are used as indices of depression. A mouse is considered to have entered an arm when all four paws are on the arm. The apparatus is cleaned thoroughly between trials with damp and dry towels. All behavioral recordings will be carried out with the observer unaware of the treatment received by mice.

Animals will be treated with *R. arboreum* extract of single dose and the effect seen will be compared with standars drugs . If extract shows depressant effect it will be recorded and compared. If it fails to exhibits anti-depression on single dose then the dosing will be continued for 21 days and test is repeated. The effect will be measured using Elevated plus – maze test model.

Experimental protocol
Animals were divided into four groups of six animals each. The groups were as follows:
Group 1: Normal control (mice administered with (normal saline 10ml/kg b.w.p.o.) daily for 21 days
Group 2: Standard *Imipramine* (animals were administered with single dose of *Imipranime* (10 mg/kg, p.o.) daily for 21 days
Group 3: Animals received single dose ethanolic extract of *R. arboreum (*200 mg/kg p.o*.)* daily for 21 days
Group 4: Animals received single dose of ethanolic extract of *R. arboreum (*400 mg/kg p.o. *)* daily for 21 days

3. RESULTS AND DISCUSSION

3.1 Extraction and Phytochemical Investigation
Successive Soxhlet extractions of *R.arboreum* was performed. The extract powder was redish brown in colour and hygroscopic in nature. The *R.arboreum* extract was subjected to different preliminary chemical tests to determine the chemical constituents present in the extract. The results has indicated the presence of flavonoids, steroids and triterpenoids, phenolic compounds and tannin as shown in Table 1.

Table 1: Test for phytoconstituents of R. arboreum flower extract

Phytoconstituents	***R. arboreum* flower extract**
Alkaloids	-

Flavonoids	+
Terpenoids	+
Anthraquinones	-
Tannins	+
Steroids	+
Phenolic compound	+
Glycosides	-
Reducing sugar	-

(+)- Present

(-)- Absent

3.2 Evaluation of Adaptogenic activity

Model I: Anoxia Stress Tolerance

The Anoxia tolerance test was determined by taking the appearance of convulsion as end point. The ethanolic extract of *R.arboreum* at low dose (i.e. EERA-I 200 mg/kg/ b.w) and high dose (EERA-II 400 mg/kg/ b.w.) showed significant ($p<0.001$) increasing tolerance stress time (i.e. onset of convulsion time) in 7^{th},14^{th} ,21^{st} day as compared with the control. Results were depicted Table 2 and graphically represented in Fig 1.

Table 2: Effect of ethanol extract of *R. arboreum* flower *on* anoxia stress tolerance test in mice

Treatment group	Dosein (mg/kg,p.o.)	Duration of Anoxia Stress Tolerance (min)			
		1st Day	7th Day	14th Day	21st Day
Control	–	20.8± 0.583	21.8±0.833	22±0.365	22.7±0.211
Standard (*Withania somnifera*)	100mg/kg	23.8±0.374**	25.8±0.749**	27.5±0.601***	32.2±0.477***
EERA-I	200 mg/kg	21.4±0.51	23.5±0.619	23.7±0.333*	25±0.365**
EERA-II	400 mg/kg	22.8±0.49*	24.5±0.428 *	24±0.365**	28.2±0.477***

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.001

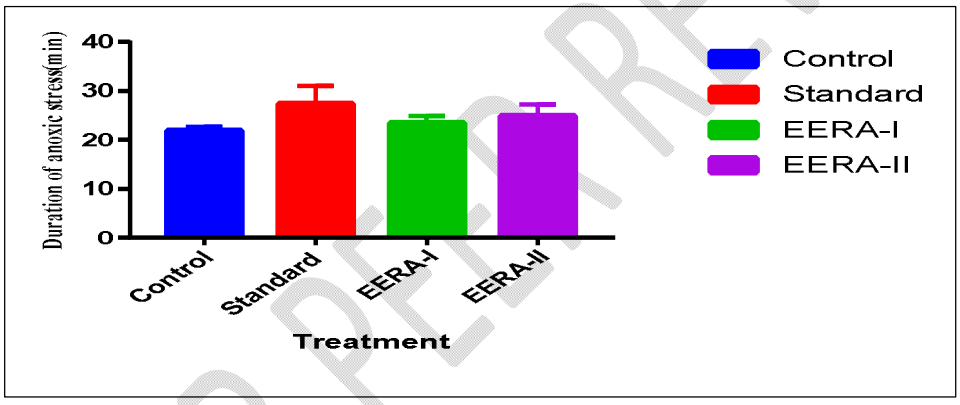

Fig.1: Effect of ethanol extract of *R.arboreum* flower on Anoxia stress test

Model II: swimming endurance test

The ethanolic extract of *R.arboreum* at low dose (i.e. EERA-I 200 mg/kg/b.w) and high dose ((i.e. EERA-II 400 mg/kg/ b.w.) showed significant(p<0. 001) increase in swimming time and is supported by estimating the biochemical parameters such as blood glucose, triglyceride, cholesterol and haematological parameter such as RBCs, WBCs and DLC. The organ weight of liver, spleen and adrenal gland was reduced in groups treated with ethanolic extract of *R.arboreum* and standard *Withania somnifera* as compared to stress control. Results were depicted Table 3, 4, 5 and graphically represented in Figure 2, 3, 4.

Table 3: Effect of ethanol extract of *R.arboreum* on swimming endurance test

Treatment group	Dose in mg/kg, p.o.	Swimming survival time (Min)

Control	–	225±2.8
Standard (*Withania somnifera*)	100	368±5.88***
EERA-I	200	240±1.83*
EERA-II	400	246±2.39**

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.001

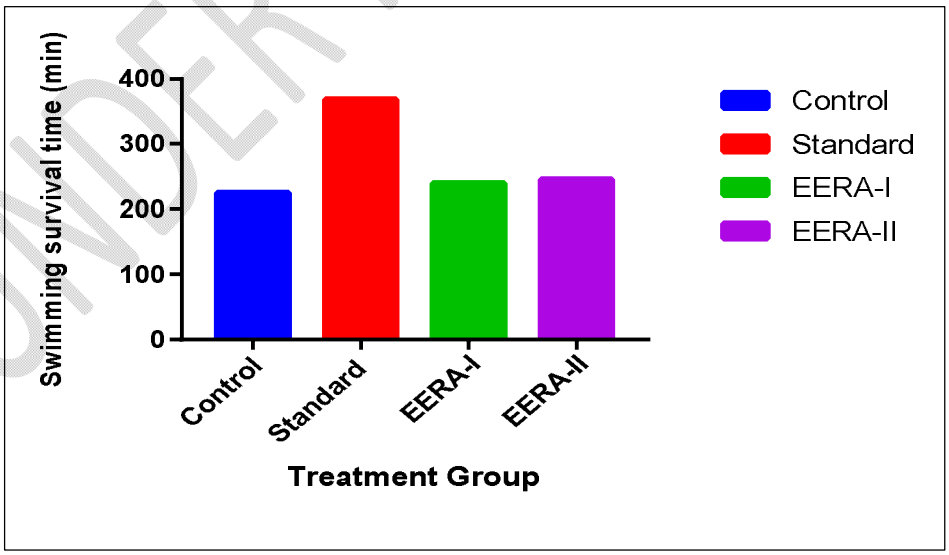

Fig. 2: Effect of ethanolic extract of *R.arboreum* flower on swimming endurance test

Table 4: Effect of ethanol extract of *R.arboreum* flower on biological parameters in swimming endurance test

Groups	Glucose(mg/dl)	Cholestrol(mg/dl)	Triglycerides(mg/dl)	BUN(mg/dl)
Control	185±4.08	49.7±0.667	53.3±0.882	37.3±0.494
Standard (*withania somnifera)*	118±4.22***	41.7±1.67**	33.2±0.872***	22.8±0.703***
EERA-I	170±2.89*	44.3±1.56*	49.5±0.619*	35.3±1.67
EERA-II	163±1.3.82**	43.7±1.56*	42±0.775**	30.8±1.4**

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.001

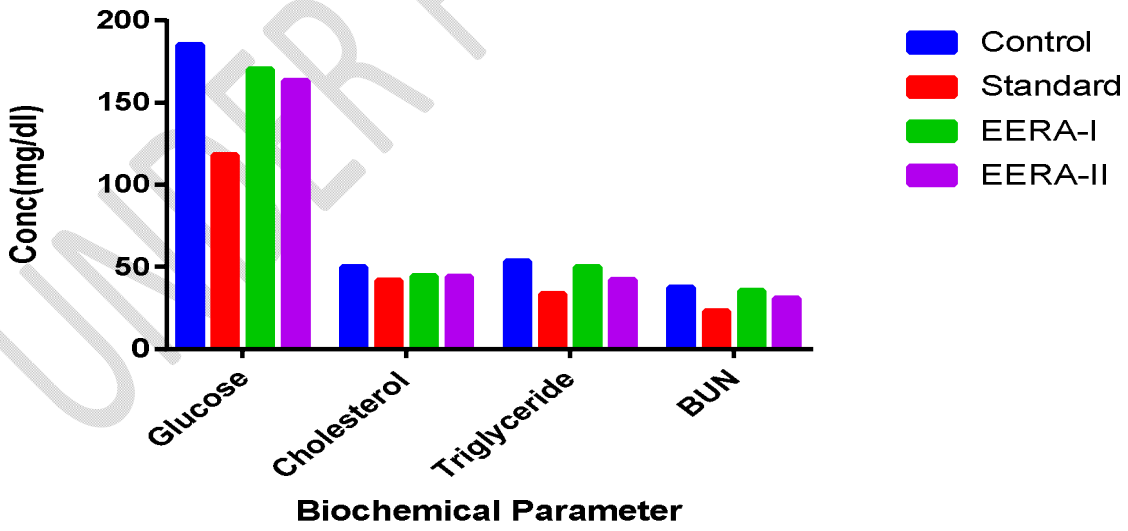

Fig.3: **Effect of ethanolic extract of *R.arboreum* on biological parameters in swimming endurance test**

Table 5: Effect of ethanol extract of *R.arboreum* flower on biological parameters in swimming endurance test

Groups	Glucose (mg/dl)	Cholestrol (mg/dl)	Triglycerides (mg/dl)	BUN (mg/dl)
Control	185±4.08	49.7±0.667	53.3±0.882	37.3±0.494
Standard (*withania somnifera)*	118±4.22***	41.7±1.67**	33.2±0.872***	22.8±0.703***
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Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett‟s. *P<0.05, **P<0.01, ***P<0.001

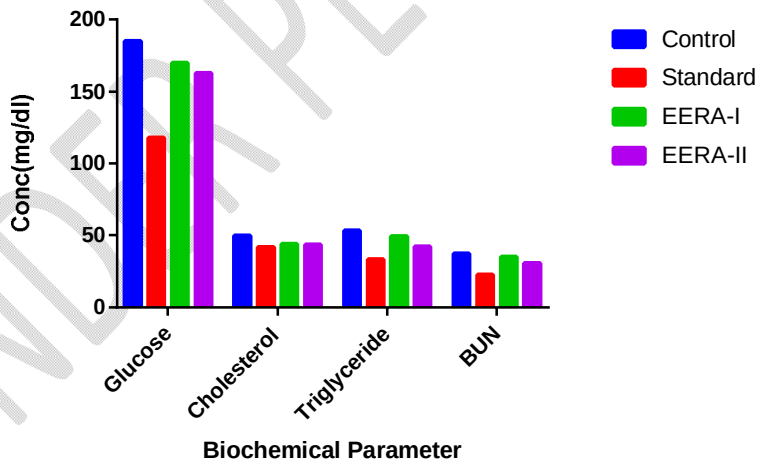

Fig.4: Effect of ethanolic extract of *R.arboreum* on biological parameters in swimming endurance test

Table 6: Effect of ethanolic extract of *R.arboreum* organ weight in swimming endurance test

Treatment group	Dose in mg/kg ,p.o.	Liver(g)	Spleen (g)	Adrenal Gland(g)
Control	-	6.6±0.13	1.09±0.0489	0.144±0.00346
Standard	100 mg/kg	3.9±0.054***	0.874±0.0246*	0.0932±0.0106**
EERA-I	200 mg/kg	5.1±0.171*	0.967±0.0989	0.112±0.00707*
EERA-II	400 mg/kg	3.88±0.215**	0.867±0.0333*	0.107±0.00516**

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.00

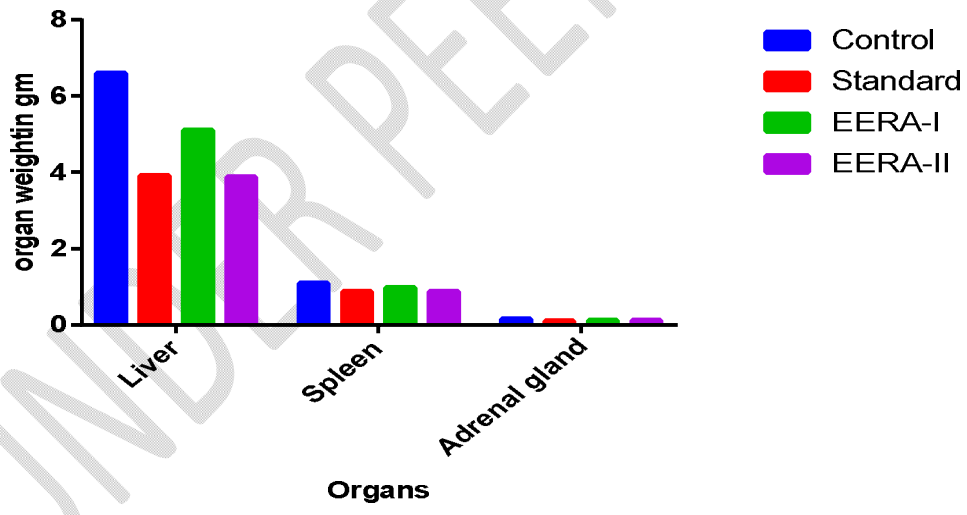

Fig. 5 Effect of ethanol extract of *R.arboreum* organ weight in swimming endurance test

Table 7: Effect of ethanol extract of *R.arboreum* on (Blood Cell Count) in swimming endurance test

Treatment group	Dose in (mg/kg .p.o.)	RBC (millions/cumm)	WBC(Cells/ cumm)	Differential Leucocytes Count (cell/cumm)		
				Lymphocytes (cell/cumm)	Neutrophils (cell/cumm)	Eosionophils(cell/cumm)
Control	–	10.7±0.441	10402±250	65.4±1.46	65.4±1,46	3.05±0.0764
Standard (*withania somnifera*)	100	8.3±0.251**	9651±143*	69.3±1.04	69.3±1.04**	2.38±0.0601**
EERA-I	200	9.69±0.396	9887±155	68.2±0.477	68.2±0.477	2.75±0.123
EERA-II	400	9.12±0.381*	9587±203*	69.2±1.28	69.2±1.28	2.72±0.0946*

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.001

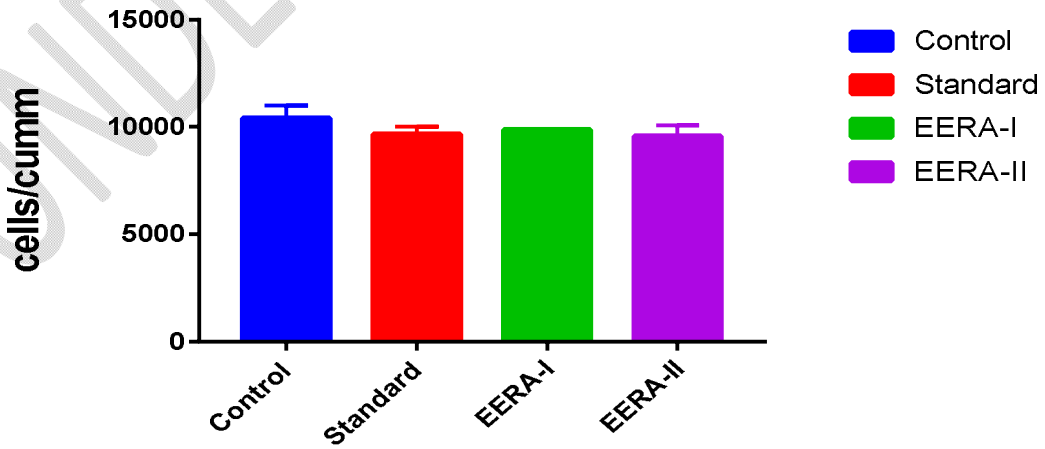

Fig.6: Effect of ethanolic extract of *R.arboreum* on WBC in swimming endurance test

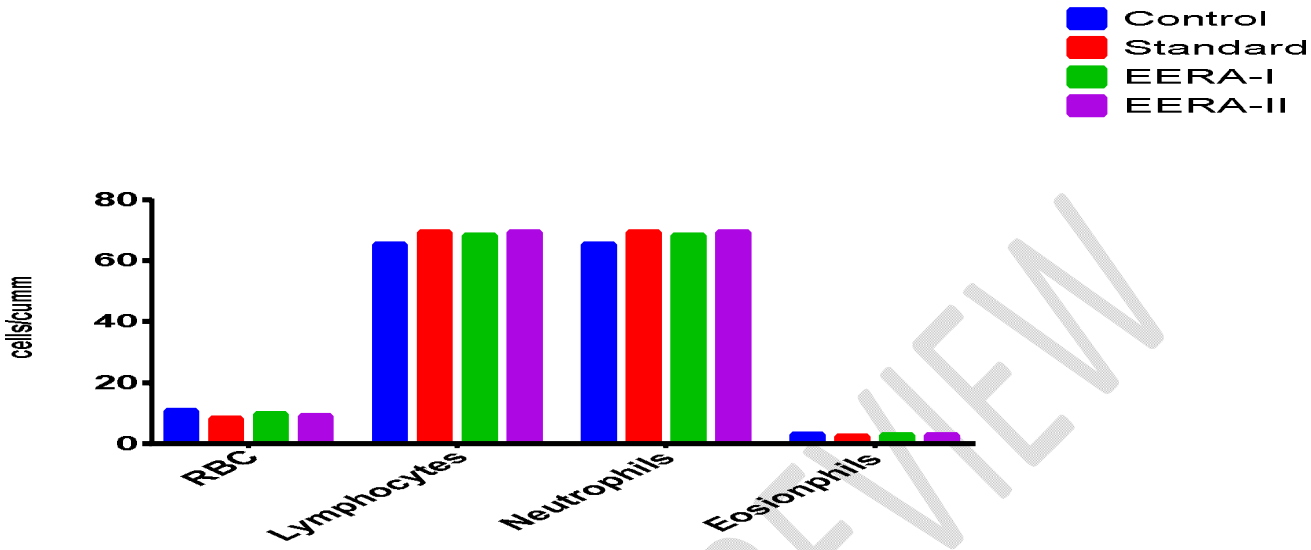

Fig.7: Effect of ethanolic extract of *R.arboreum* on (Blood Cell Count) in swimming endurance test

3.3 Tail Suspension Test

Antidepressant activity of ethanolic extract of *R.arboreum* was studied in mice by tail suspension test. The duration of immobility was reduced after treated with ethanolic extract of *R.arboreum as* compared to control. The effect of ethanolic extract of *R.arboreum* at low dose (200 mg/kg, p.o.) and high dose (400mg/kg,p.o.) was near to standard Imipramine. Study indicates that ethanolic extract at both dose (i.e.200 mg/kg, p.o and 400 mg/kg, p.o.) has considered as significant antidepressant activity. Results were depicted Table 8 and graphically represented in Figure 8

Table 8: Effect of ethanol extract of *R. arboreum* on tail suspension test

Treatment group	Dose in mg /kg (p.o.)	Duration of immobility(sec)		
		1st day	7th day	14th day
Control	-	239±1.83	236±3	231±2.39
Standard (*withania somnifera*)	100mg/kg	232±1.3*	223±2.14**	213±2.14***
EERA-I	200mg/kg	236±01.53	230±1.54	217±2.19**
EERA-II	400mg/kg	234±1.73	225±1.86**	216±2.39***

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.001

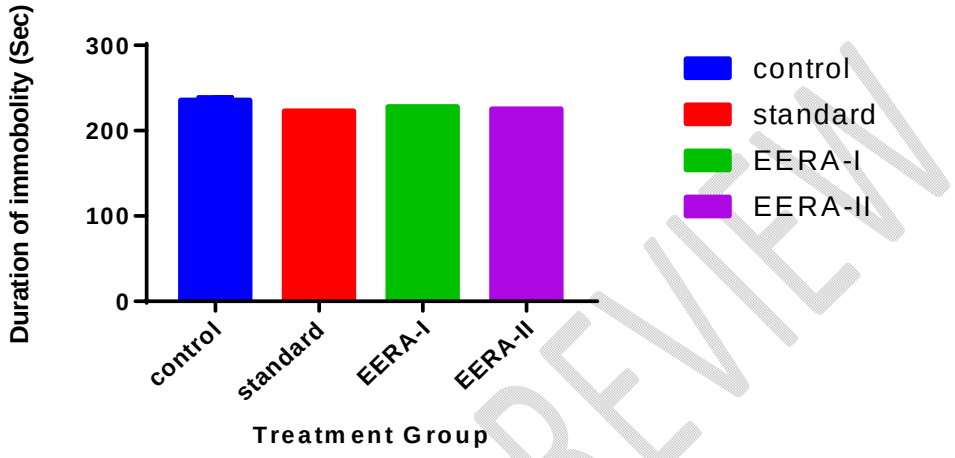

Fig.8: Effect of ethanolic extract of *R.arboreum* flower on tail suspension test

3.4 Elevated Plus Maze Test

According to the result (Table no.9) the time spend and number of entries in open arm were significantly increased in high dose and standard (figure no.9 & 10) and there was significantly decreased in time spend and number of entries in enclosed arm (figure no.9 & 10) as compared to control. The study result (table no.9) showed that the *R. arboreum* flowers extract in high dose significantly increase in the exploration time and entries in to open arm. It shows increase in the mobility of the mice in open arm which suggest the antidepressant activity.

Table 9: Effect of ethanolic extract of *R. arboreum* on Elevated plus maze test model mice

Group	Treatment	Time spend in open arm (Sec.) Mean ± SEM	Time spend in enclosed arm (Sec.) Mean ± SEM	No. Of entries in open arm Mean ± SEM	No. Of entries in enclosed arm Mean ± SEM
Control	Saline	45.2± 4.91	254 ± 5.01	5.67 ± 0.333	10.20 ± 0.477
Standard	Imipramine (10mg/kg)	66.5± 3.91**	233 ± 3.81 **	10 ± 0.577 **	7.83 ± 0.601 *
Low Dose	EERA(200mg/kg)	54.2± 3.52	244 ± 3.99	7.17 ± 0.601	8.83 ± 0.601
High Dose	EERA(400mg/kg)	60 ± 3.65 *	239 ± 12.81*	8.17 ± 0.601*	7.67 ± 0.558 *

Results are expressed as mean ±SEM(n=6), data analyzed by using one way ANOVA followed by Dunnett"s. *P<0.05, **P<0.01, ***P<0.

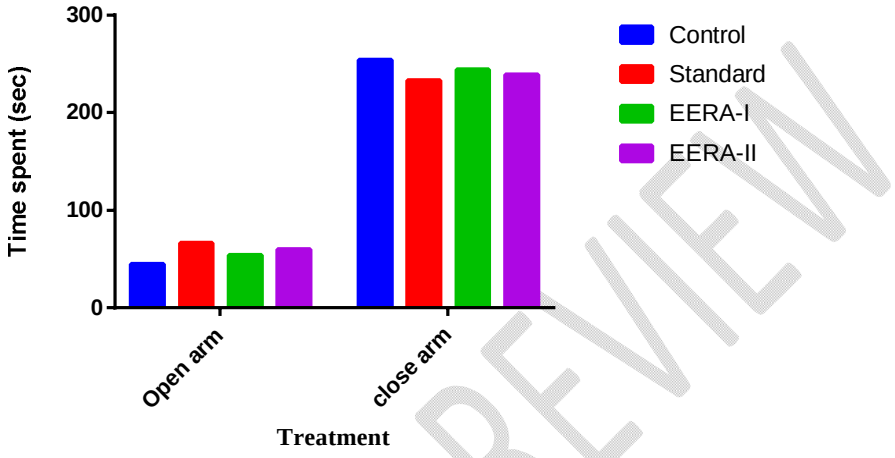

Fig. 9: Effects of ethanolic extract of *R. arboreum* on Time spend in open arm & in enclosed arm.

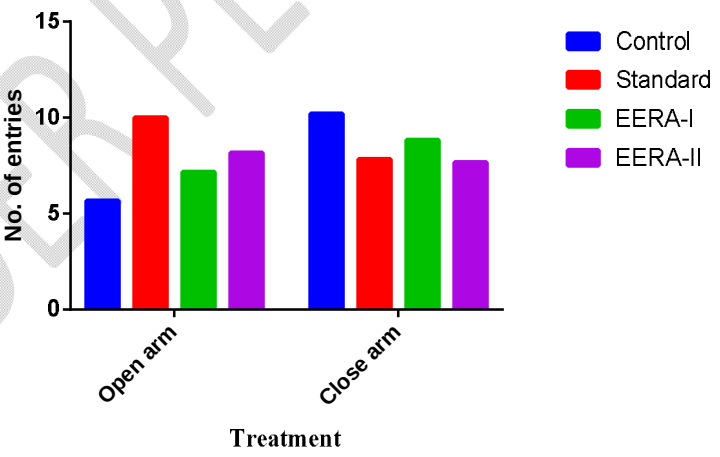

Fig. 10: Effects of ethanolic extract of *R. arboreum* on number of entries in open arm & entries in enclosed arm

DISCUSSION

In The preliminary phytochemical screening of the flower extracts of R. arboreum Sm. Ssp. Nilagiricium (Zenker) Tagg carried out by Kiruba and co-worker showed the presence of various bioactive compounds such as phenols, saponins, steroids, tannin, xanthoprotein and coumarin. These compounds have been reported to possess several pharmacological properties such as antioxidant, anti-inflammatory, antimicrobial, and anticancer activities. whereas on my current study it was found the presence of flavonoids, steroids and triterpenoids, phenolic compounds, and tannins in the flower extracts of R. arboreum Sm. ssp. nilagiricum (Zenker) Tagg. The presence of flavonoids and phenolic compounds is particularly noteworthy as these are well-known for their antioxidant properties and can prevent or delay the onset of chronic diseases

In another study, Mohammad Nisar and co-worker investigated the antibacterial and cytotoxic activities of various parts of the R. arboreum plant extract, including the flower, leaves, bark, stem, and roots. The results showed significant antibacterial activity against medically important pathogens such as Salmonella typhi, E. coli, Staphylococcous aureus, and Bacillus subtilus. The cytotoxicity of the crude extract was also found to be effective against A. salina at a concentration of 1000µg/ml [14, 15].

In the various studies conducted on Rhododendron arboretum ,Neeraj et al. found that the ethyl acetate fraction of Rhododendron arboreum flowers had a potent hepatoprotective effect against carbon tetrachloride-induced hepatic damage in rats. They suggested that this effect was due to glutathione-mediated detoxification and free radical scavenging activity. Sonar et al. investigated the anti-microbial and phytochemical properties of Rhododendron arboreum flowers. They found that the alcoholic extract of the plant was more active against bacterial strains than fungal strains. They suggested that the observed anti-microbial activity was due to the presence of Quercetin and other bioactive agents. and In another studies conducted by Roy et al. a on the leaves of Rhododendron arboreum found that the plant extract possessed potent anti-stress activity in test animals. They concluded that the adaptogenic activity was due to the presence of strong antioxidant activity from flavonoids and gallic acid [16-18].

Our study conducted on the ethanolic extract of Rhododendron arboreum to evaluate its **anoxia tolerance, swimming endurance, and antidepressant activity**. The study found that the extract exhibited significant improvements in tolerance stress time, swimming time, and exploration time in open arms in mice. Additionally, the extract showed a noteworthy reduction in the duration of immobility in the tail suspension test, suggesting its potential as an antidepressant agent.

Overall, these studies suggest that Rhododendron arboreum may possess various pharmacological properties, including hepatoprotective, anti-stress, anti-microbial, anoxia tolerance, and antidepressant activities. However, further studies are needed to confirm these findings and explore the underlying mechanisms of action.

The present study was carried out in healthy animals to analyze the adaptogenic and antidepressant activity. The detailed study should be conducted in the human volunteers who are suffering from depression and stress.

CONCLUSION

The present study suggests that the ethanolic extract of *R. arboreum* flower has significant adaptogenic and antidepressant activity. The biochemical, haematological and organs weight in rat also support adaptogenic activity of ethanolic extract of *R. arboreum* flower. This adaptogenic activity in title plant might be due to presence of flavonoids like rutin and Quercetin and Antidepressant activity might be due to terpenes. However, further investigation should be carried out to elucidate the exact mechanism of action.

CONSENT

It is not applicable

ETHICAL APPROVAL

The study protocol was approved by the Institutional Animal Ethics Committee (IAEC), Office of institutional animal ethical committee (IAEC) of Mallige College of pharmacy, Banglore. Reg. no. 1432/PO/Re/S/11/CPCSEA.

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