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# **ANXIETY PROTECTIVE ACTIVITY OF FICUS RACEMOSA ON SWISS ALBINO MICE**

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**OBJECTIVE:** To perform anti-anxiety properties of hydro-methanolic extract of ficus racemosa (leaf) on experimental models (elevated plus maze and Open- Field test) in by using mice.

**MATERIALS AND METHODS:** Leaves of ficus racemosa was collected, dried and crushed. The leaves were extracted with hydro-methanolic by using soxhlet method of extraction. After the extraction phyto-chemical screening were PERFORMED. The hydro-methanolic extract leaf of ficus racemosa was studied at dual dosage level (200mg/kg and 400mg/kg, in p.o. administration) in mice by applying EPM and OFT model. The animals are shared into 4 sets and every group contain 6 mice. Group-I mice were managed using normal saline for 14 days. Group-II mice were managed by standard drug (Diazepam) 1mg/kg for 14 days. Group-III mice were managed by excerpt of f. racemosa at a dosage of 200mg/kg and Group-IV mice were managed with excerpt of f. racemosa at a dosage 400mg/kg for 14 days. Anti-anxiety action of hydro-methanolic extract of f. racemosa was estimated by EPM model and Open- Field apparatus test. In EPM and OFT test, aqueous extract of f. racemosa showed increased the % of time spent in open arm and % of open arm entries.

**RESULTS & DISCUSSION:** Phyto-chemical investigation of f. racemosa aqueous excerpt exposed the occurrence of alkaloids, flavonoids, amino acids, proteins, and carbohydrates. Administration of Diazepam (1mg/kg i.p.) produced a significantly decrease the anxiety behavior. when matched by control treated group. Admin of f. racemosa excerpt (200mg/kg & 400mg/kg) to mice significantly decrease the anxiety like behaviour when matched through Diazepam cured group and it was found that abstract dose 400mg/kg show higher anxiolytic effect than 200mg/kg dose of excerpt of ficus racemosa.

**Conclusion:** Results suggest that leaves of f. racemosa possess significantly anti-anxiety activity.

**KEYWORDS:** Ficus racemosa, Diazepam, Anti-anxiety, EPM, OFT.

## **I. ANXIETY**

Anxiety is a mental illness that throws off our everyday routines, including motivation and mood. Severe anxiety leads to mental illness and anxiety disorders, which are typically treated with synthetic drugs. Unease, discomfort, and worry or fear about a recognized threat are the hallmarks of anxiety, a negative emotional state. Anxiety is a typical occurrence in life. Treatment is necessary when it is excessive and out of proportion to the circumstances. Anxiety conditions are the utmost mutual mental health disorders. It shows itself as irregularities in our physiological activity, emotions, thoughts, and behavior. Acute stress condition, obsessive-compulsive condition, social anxiety, fear of heights, generalized anxiety disorder, particular phobia, panic attack condition, and post-traumatic stress disorder are a few kinds of anxiety conditions..[1] Anxiety has been associated for at least six months with impatience, restlessness, a tight or tense feeling, a tendency to get tired easily, difficulty focusing or losing focus, difficulty falling asleep, and irritation. [2] Anxiety is among the utmost communal mental and behavioral issues.



**Figure no. 1. Anxiety condition.**

## **1.1 Causes of anxiety disorder:**

### **1.1.1. Biological factors:**

- Heredities
- Neurotransmitter disorder
- Sickness
- Drugs
- Dietary habits

### **1.1.1. Psychological factors:**

- Behavioural characteristics
- Poor self-worth
- Unfavourable feelings
- Perceptions of surroundings

### **1.1.2. Social factors:**

- Negative circumstances in life,
- A lack of interaction with others
- Stress in the workplace
- A lack of social abilities
- Events of nature

## **1.2. Kinds of anxiety disorders:**

### **1.2.1. Generalized Anxiety Disorder (GAD):**

Unnecessary stress and worry over a variety of measures or actions that occur more often than not throughout a minimum of 6 months, along through the signs that go along with it (e.g., exhaustion, poor concentration).

### **1.2.2. Specific Phobia:**

Pronounce and ongoing fear of things or circumstances that are easily identifiable (e.g., flying, heights, animals).

### **1.2.3. Post Traumatic Stress Disorder:**

Anger in reaction to a harrowing event in which bodily damage happened or was in danger (such as war, or a natural disaster, child misuse rape), flashbacks, or recurring frightening thoughts and recollections.

**1.2.4. Social Phobia:** Social anxiety condition is another name for it. An immediate anxiety reaction is nearly always triggered by social or performance settings. [3].

**1.2.5. Obsessive-Compulsive Disorder Obsessions:** Judgments, planning, urges, or pictures that are persistent, inappropriate, and create a great deal of concern or suffering.



**1.2.6. Compulsions:** Repetitive actions (like washing your hands, comparison, or testing) or mental processes that are triggered by rituals or obsessions.

**1.2.7. Panic disorder:** The existence of frequent, unplanned calm attacks, monitored via a major shift in manners linked to attacks, or near about 1 month of ongoing worry around having more attacks or implications of its repercussions. [4].

### 1.3. Signs and symptoms of the anxiety disorder:-

- Sense agitated
- Being certainly tired
- Headaches, muscle aches, stomach aches or unexplained pains
- Sweating
- Chest pain
- Being out of control
- Difficulty making eye contact to people they don't know
- Having sleep problems
- Difficulty in controlling to feeling of worry
- Difficulty in concentrating
- Racing heart.[5]

## II. MATERIAL AND METHODS

### 2.1. Collection of plant materials:-

Plant was picked for proposal view of its customary, phytochemical profile was assembled from herbal garden of R.K. Pharmacy College in Kashipur, Surai, Sathion, Azamgarh, Uttar Pradesh.

### 2.2. Authentication of plant:-

Prof. N. K. Dubey, Section of Botany, Banaras Hindu University Varanasi, India, approved the plant. For future reference, a voucher example no Mora. 2025/01 was set at the herbarium of BHU Varanasi.

### 2.3. Requirements:-

**Table No. 1: Requirements:-**

Instruments	Chemicals	Drug
Digital pH meter, Soxhlet extractor	Mayer's, Wagner's, Shinoda	Diazepam
Heating mantle, Condenser	Hager's, Lead acetate	
Desiccator, Grinder	Biuret, Ninhydrin, Foam test	
EPM	Ferric chloride test	
OFT apparatus	Keller-killiani test, Benedict test	
Syringe	Molisch's test reagents	
Oral cannula	Methanol Reagents and Distilled Water	

### 2.4. Preparation of extracts:-

Coarse Powdered (300g) leaf of plant was defatted with hydro alcoholic (methanol) ,(60-80° C), and then completely removed with methanol using soxhlet apparatus at 60°C for eight hour. Extract was sifted on whatman No. 1 filter paper. Solvent was completely removed under reduced pressure in a rotator vacuum evaporator to get a powdery mass. The concentrated extract was stored in vacuum desiccators for further use. HME was tried for anti-anxiety properties by quantity stages of 200 & 400 mg per kg orally. The results were matched by standard antianxiety agent diazepam (1mg/kg i.p.). All the test materials were suspended in vehicle (water)[6].

## 2.5. Experimental Animals:-

For this experiments utilize swiss albino mice balancing 25-30 gms. In polypropylene cages (38 x 23 x 10 cm) with paddy husk as bedding was arrangement for all rats treatment groups. Organization Temperature of 25±2°C, comparative humidity of 50±5 percent rats housed. Monitored light (12 h dim / 12 h sunny period). Fresh water ad libitum, fed using normal pellet nutrition allowed to all animals. The experiments was performed following animals ethics guidelines of “Institutional Animals Ethics Committee” (Approval No: 1384/PO/Re/S/10/CCSEA) and project id no. IAEC/RK/25/06.

Table No. 2 Grouping of animals

S. No.	Groups	No. of Animals
1	Normal Control normal saline) P.O.	6
2	Standard (Diazepam) 1 mg/kg I.P.	6
3	FRHME 200 mg/kg P.O	6
4	FRHME 400 mg/kg P.O.	6

## 2.6. Elevated plus maze induced anxiety model:

The antianxiety activity of a hydro-methanolic excerpt of plant was studied in Swiss albino mice using an elevated plus-maze approach. The plus maze was shaped like a cross or plus, through double open (30 x 5 cm x 0.2 5 cm) and double closed (30 cm x 5 cm x 15 cm) arms stretching from centre boards (5 cm x 5 cm) and rising to 50 cm beyond the ground. The whole maze was prepared of wood. Animals balancing 20-30 grams were put into 4 groups of every six.

Group-I is control administered only normal saline, group-II standard medication (Diazepam 1mg/kg) was given intra-peritoneal, group-III (Test-I) plant extract 200mg/kg and group-IV (Test-II) 400mg/kg orally was given. Standard I.P. was administered 30 minutes beforehand analysis, and extracts were administered P.O. 45 minutes prior to experiment. Animals were engaged individually in the epicentre of maze, facing an open arm, and number of entrances and time spent in closed and open arms were noted for a 5-minute remark period. Arm entrances were defined as all four paws entering in arm.[7].

Group-I is control administered only normal saline.

Group-II standard medication (Diazepam 1mg/kg) was given I.P.

Group-III (Test-I) FRHME 200mg/kg.

Group-IV (Test-II) FRHME 400mg/kg orally was given

All treatment for given 14 days.

## 2.7. Open- Field apparatus test (OFT):

Albino mice (25-30 g) of both gender were engaged into four crowds of six mice every one. Previous to test, the creatures were abstained overnight but given free admittance to water. Group I was treated as a normal control, receiving vehicle once every day for 14 days; Group II was given diazepam (1 mg/kg, I.P.); and Groups III and IV were given varying dosages of test excerpts P.O. once every day for 14 days. On the 14th day, 1 hour afterward administering the vehicle, standard, and test extracts, every animal was engaged in centre of an open field ground, and subsequent parameters were noted through a 5-minute test[8].

**Ambulation:** Measuring the amount of squares crossed via mice.

**Rearing:** The number of times the mice stood on its hind legs.

Group-I is control administered only normal saline.

Group-II standard medication (Diazepam 1mg/kg) was given i.p..

Group-III (Test-I) FRHME 200mg/kg.

Group-IV (Test-II) FRHME 400mg/kg orally was given.

All treatment for given 14 days

**2.8.. Statistical Analysis:** Statistical Analysis: The results were presented as mean  $\pm$  SEM. The variances were matched utilizing one-way ANOVA and Dunnett's test.

### III. RESULT

#### 3.1. Phytochemical Screening:-

*F. racemosa* exposed the existence of tri-terpenoids, proteins and amino acids, alkaloids, flavonoids, and coumarins. In order to address the existence of various phenolic mixes, such as polyphenols, flavonoids, phenolic acid, etc.

**Table No. 3:** Preliminary phytochemical investigation.

Phytochemicals	Name of the test	Observation	Result
Alkaloids	Mayer's	Creamy white ppt.	+
	Wagner's	Deep chocolate ppt.	+
	Hager's	Yellow crystalline precipitate	+
Flavonoids	Shinoda	Development pink colour	+
	Specific	Orange to red	+
	Lead acetate	Yellow coloured	+
Proteins and Amino acids	Biuret	A pink coloured	+
	Millon's	A white precipitate develop	+
	Ninhydrin	No precipitate	-
Glycosides	General test Legal's test	Yellow pigment	+
	Baljet test	Pink to red pigment	+
		No ppt	-
Tri-terpenoids	Salkowski's test	Develop Golden yellow layer	+
Tannins	Gelatin test	No precipitate	-
Steroid	Liebermann-Burchard's test	No precipitate	-

#### 3.2. Effect of *ficus racemosa* leaf excerpt on Elevated Plus-maze Model:

The hydro-alcoholic excerpts of *ficus racemosa* leaf at dosages of 200 mg/kg and 400 mg/kg p.o. completed well in EPM (Table 4). It significantly improved the number of entries and the time spent in the open arm. The potency of the anti-anxiety action is 400mg/kg p.o. The pharmacokinetics of hydro-methanolic excerpts of *ficus racemosa* were comparable to diazepam 1 mg/kg i.p.

**Table no. 4** Anxiolytic activity of hydro-methanolic excerpt of *ficus racemosa* plant on mice epm test:

S. no.	Group	Treatment	Number of entries open arm (counts)	Number of entries closed arm (counts)	Time spent in open arm (s)	Time spent in closed arm (s)
1	Group-I	Control (normal saline)	5.43 $\pm$ 0.42	14.33 $\pm$ 0.55	67.5 $\pm$ 0.56	230.4 $\pm$ 0.46
2	Group-II	Diazepam(1mg/kg)	9.60 $\pm$ 0.55*	6.33 $\pm$ 0.66*	180.54 $\pm$ 0.67*	117.20 $\pm$ 0.57*
3	Group-III	HMEFR(200mg/kg)	6.68 $\pm$ 0.11ns	9.56 $\pm$ 0.33*	92.21 $\pm$ 0.60*	205.72 $\pm$ 0.40*
4	Group-IV	HMEFR(400mg/kg)	7.68 $\pm$ 0.22b	8.33 $\pm$ 0.44*	134.11 $\pm$ 0.18*	163.73 $\pm$ 18*

Values described as Mean  $\pm$  SEM (n=6). The data were examined via one-way ANOVA tracked via and Dunnett's test a-P <0.05, b- P<0.01, #- P<0.001; \*- P<0.0001; ns- non significant when matched through control group.

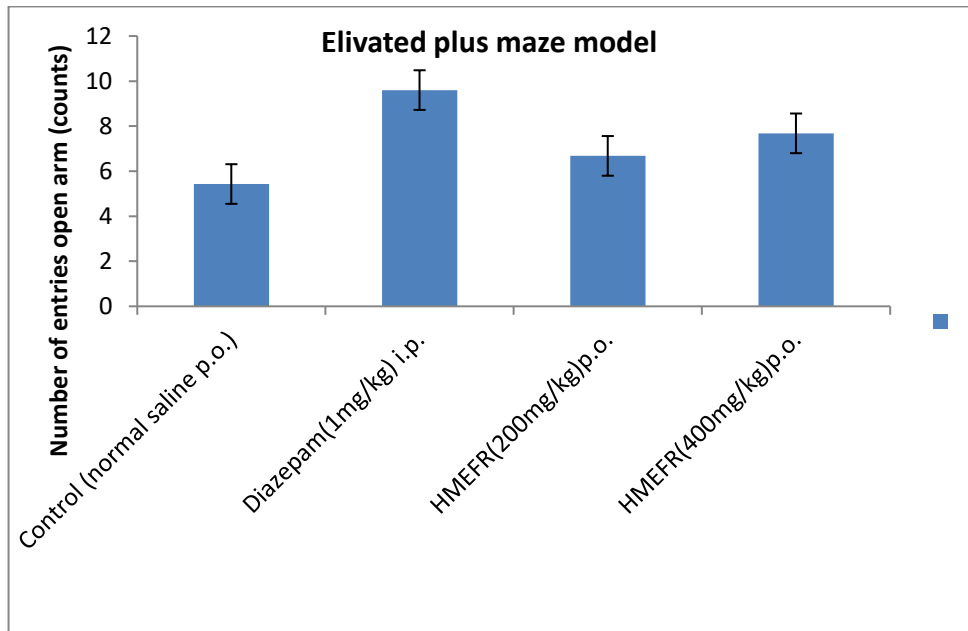


Figure no. 2. Result of EPM test of HMEFR leaf Number of open arm entries (counts):

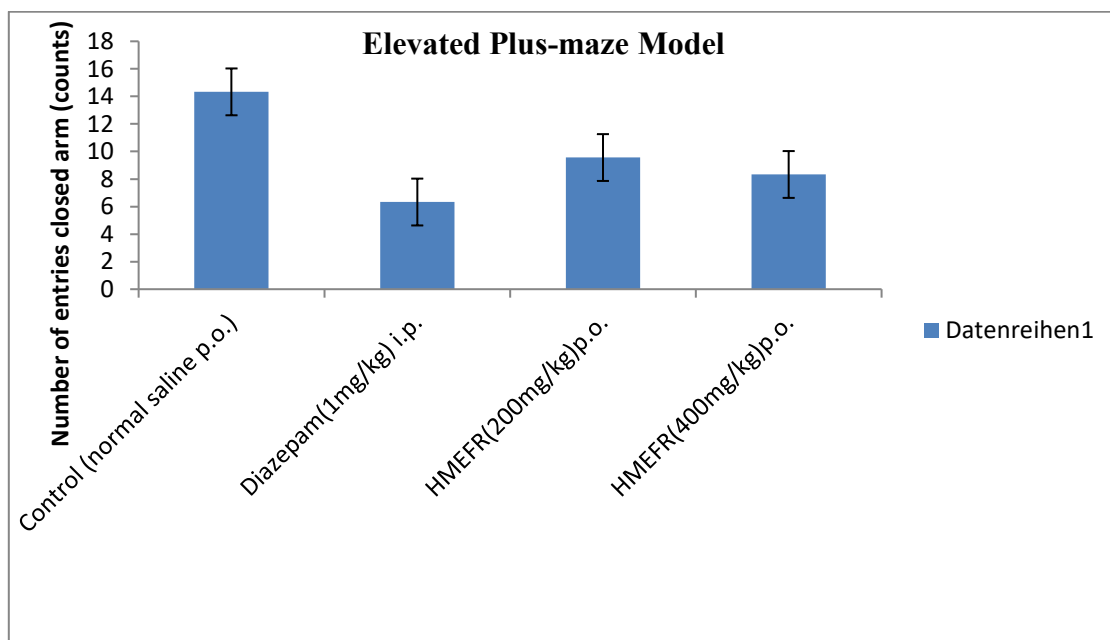


Figure no. 3. Result of EPM test of HMEFR leaf No. of closed arm entries (counts):

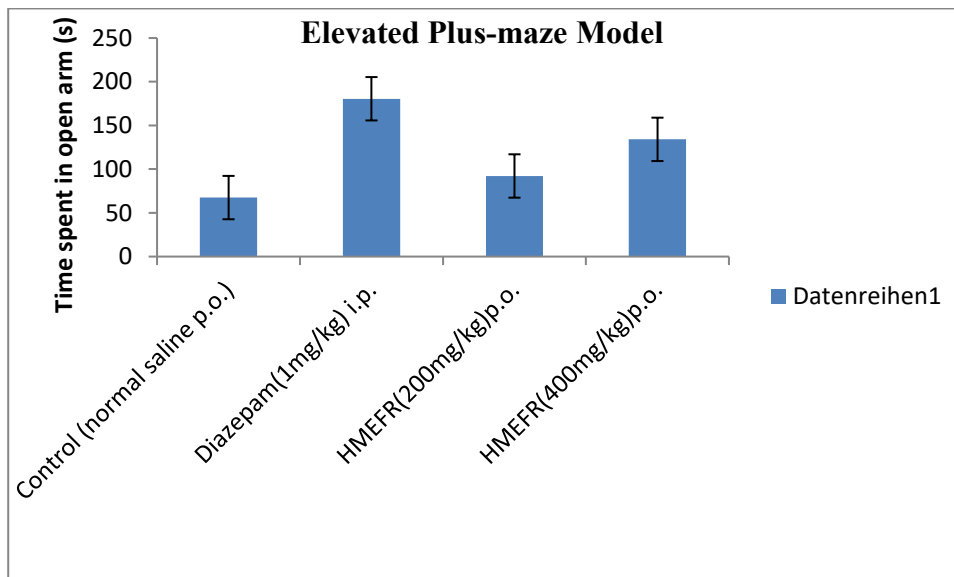


Figure no. 4. Result of EPM test of HMEFR leafs Time spent in open arm (s):

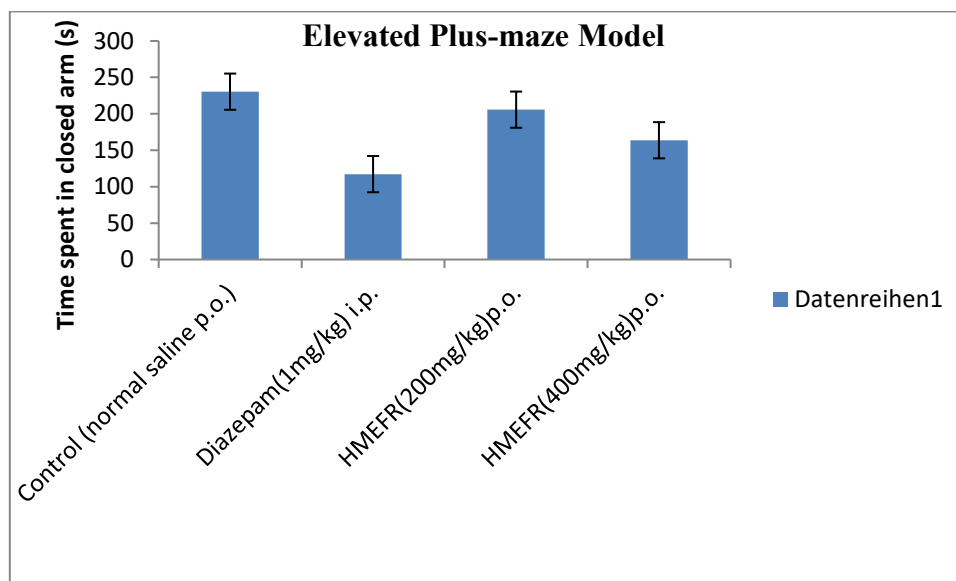


Figure no. 5. Result of EPM test of HMEFR leaf Time spent in closed arm (s):

### 3.3. Effect of ficus racemosa leaf extract on open field test:

In the open field test, diazepam and plant extract (Ficus racemosa methanol extract 200mg/kg and 400 mg/kg p.o.) cured mice displayed significant rise in the number of rearings and number of squares crossed through five min intermission as matched to vehicle treated control group. Whereas, test extract at 400 mg/kg p.o. displayed statistically significant results [Table no.5.]



Table no.5. : Effect of ficus racemosa leaf extract on the behavior of animal in open field test:

S. no.	Group	Treatment	No. of squares crossed	Rearing
1	Group-I	Control (normal saline)	120.37 $\pm$ 1.213	10.27 $\pm$ 2.69
2	Group-II	Diazepam(1mg/kg)	176.21 $\pm$ 1.11***	28.69 $\pm$ 3.64***
3	Group-III	HMEFR(200mg/kg)	135.37 $\pm$ 1.54 <sup>ns</sup>	16.49 $\pm$ 4.03 <sup>ns</sup>
4	Group-IV	HMEFR(400mg/kg)	163.08 $\pm$ 2.22***	25.51 $\pm$ 2.55**

Values are Mean SEM, (n=6), ns Non-significant\* p < 0.05, \*\* p < 0.01, \*\*\* p < 0.001 as matched to control.

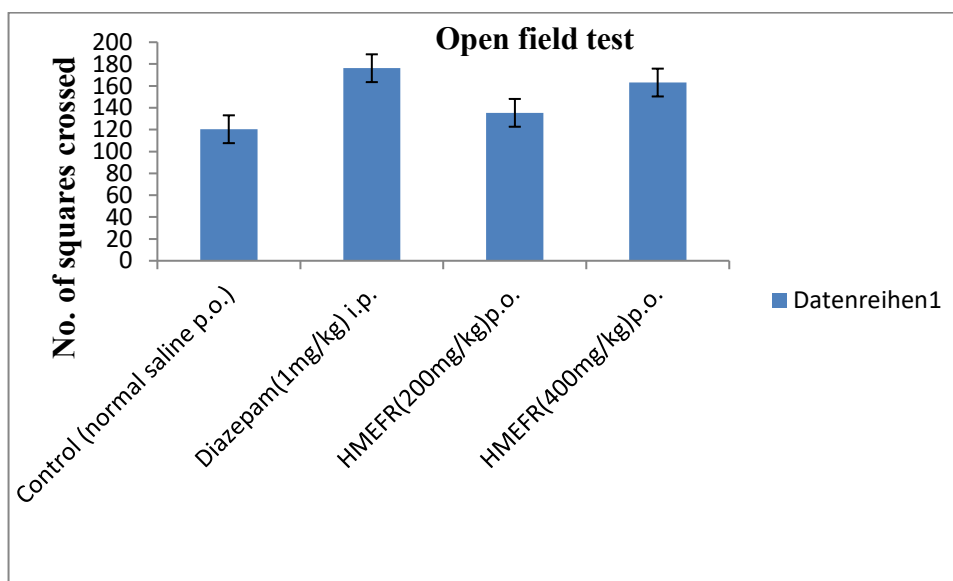


Figure no. 6. Result of OFT test of HMEFR leaves No. of squares crossed:

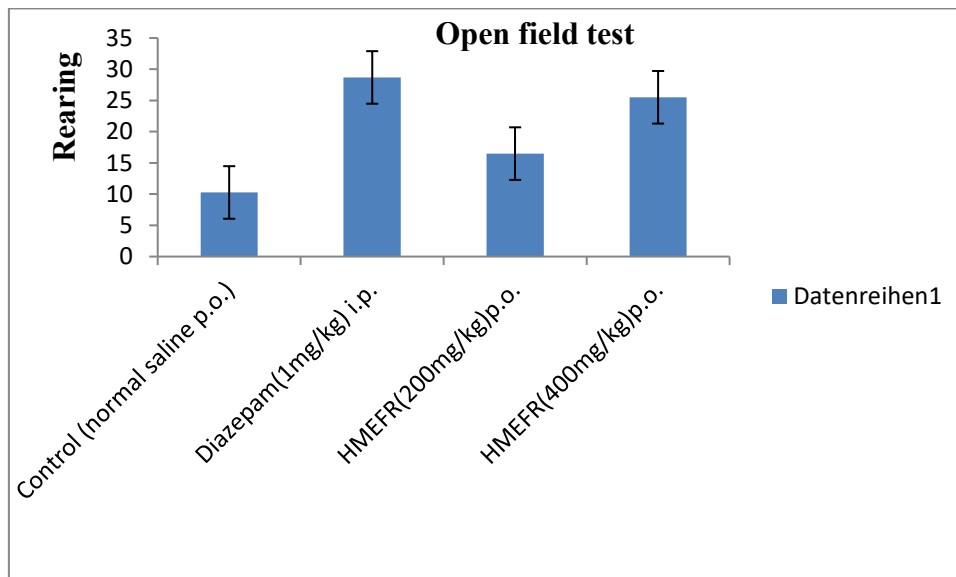


Figure no. 7. Result of OFT test of HMEFR leaves Rearing:

#### IV. DISCUSSION

Here are not any reports of scientific assessment of ficus racemosa leaves of antianxiety properties, although its historic usage for curing a variation of disorders. Alkaloids, flavonoids, proteins, and glycosides chemicals are all confirmed by the phytochemical screening. Mice were selected for the assessment of antianxiety action in experimental method since mice demonstrate robust anxiety-like behavior while showing to stressors (for example, novelty, intense light, or social conflict). [71]. The elevated plus-maze models are utilized to examine mice's psychomotor and affective features. [72]. However, the anxiolytic activity of the methanolic excerpt of f. racemosa leaves (200 and 400 mg/kg oral) was tested using EPM, which is recommended when the experiment agent rises open arm entries lacking changing the overall number of arm entries. Standard drug (diazepam) has long been used as a antianxiety, and it is as well commonly used in behavior pharmacology as a reference chemical for potentially antianxiety compounds. However, mice treated with 400 mg/kg body wt. of plant extract displayed a significant rise in the proportion of entries into open arms as well as time spent in the open arms of labyrinth. The OFT is utilized to determine the mice emotive condition. The open field model investigates anxiety-related manners defined via the animal's usual aversion to an open, light space. Thus, when creatures are removed from their adjusted cages and engaged in an unfamiliar habitat, they exhibit worry and dread by altering all or some parameters. Anxiolytic therapy lowers the scared manners of creatures in an open field. Statistical examination of records got since this trials stayed antianxiety action of plant excerpt as its influence displays significant rise in number of rearings, and number of squares crossed, as matched to vehicle-cured group, which shows its antianxiety properties. [73].

#### V. CONCLUSION

Rendering to the findings, hydro-alcoholic excerpts of ficus racemosa leaf had anti-anxiety properties. These findings suggest that the hydro-methanolic excerpt of ficus racemosa leafs has a stronger anti-anxiety effect compared to control. Additionally, allopathic drugs employed in its therapy are linked to side effects that worsen human health. Researchers are now concentrating on herbal plants that have therapeutic properties as a result of this. These medicinal plants comprise a variety of phytochemicals, containing alkaloids, flavonoids, phenols, glycoside and others; these substances can be isolated and used to promote health. Consequently, the medicinal plants with anxiolytic effect that are not only secure but also reasonably priced are explored here.

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