World Journal of Biology Pharmacy and Health Sciences
eISSN: 2582-5542
Cross Ref DOI: 10.30574/wjbphs
Journal homepage: https://wjbphs.com/

(RESEARCH ARTICLE)

Neuropharmacological study of *Moringa olifera*

Poulami Sadhu [1] and Arabinda Nayak [2,*]

[1] *Burdwan Institute of Pharmacy, Burdwan, West Bengal, India.*
[2] *Gupta College of Technological Sciences, Asansol, West Bengal, India.*

World Journal of Biology Pharmacy and Health Sciences, 2023, 14(03), 092–104

Publication history: Received on 24 April 2023; revised on 10 June 2023; accepted on 13 June 2023

Article DOI: https://doi.org/10.30574/wjbphs.2023.14.3.0237

Abstract

Objective: *Moringa olifera* is a medicinal plant widely used in many parts of the world for hypertension, pain and epilepsy. The aim of this study was to determine the effect of *M. olifera* aqueous leaf, root and steam extract on neurobehavioral activities of healthy mice.

Method: Male mice were randomly divided into a control group (given 0.9% NaCl orally) and three groups treated with the extract at doses of 100, 200, and 400 mg/kg/day for 14 consecutive days. Neuropharmacological activity were evaluated by neuro-muscular coordination (rota rod treadmill), pain (hot plate and acetic acid-induced abdominal constriction). Administration of *M. olifera* extract had a significant and dose dependent antinociceptive action in both thermal and chemical tests ($p < 0.05$). The extract (400 mg/kg) caused a reduction in exploration activity and neuromuscular coordination, and decreased the mobility time in the FST, suggesting an antidepressant-like action. Motor activity was not significantly affected by any of the doses used.

Conclusion: The results suggest that the plant aqueous extract may have a dose-dependent central nervous system (CNS) depressant action.

Keywords: Acetic acid; Hot plate; *Moringa olifera*; Neurobehavioral activities; Rota rod

1. Introduction

Basic plan of nervous system in man and other vertebrates is more or less the same.

The nervous system consists of two main parts. Central Nervous System (CNS) and Peripheral Nervous System (PNS).

1.1. Central Nervous System (CNS)

It includes the brain and spinal cord. It is the site of information processing and control. The brain is the upper part which lies enclosed in the skull in the head region, and the spinal cord which is long and narrow part which lies enclosed in the skull in the head region, and the spinal cord which is long and narrow part enclosed within the vertebral column.

1.2. Peripheral Nervous System (PNS)

It comprises all the nerves of the body associated with the CNS (brain and spinal Cord).

- Somatic Nervous System: This is the voluntary part of peripheral nervous system. It conducts impulses to and from the non-visceral components of the body like voluntary (skeletal) muscles, bones, ligaments, joints, skin receptor and peripheral sense organs.

* Corresponding author: Arabinda Nayak

Copyright © 2023 Author(s) retain the copyright of this article. This article is published under the terms of the Creative Commons Attribution Liscense 4.0.

- Visceral Nervous System: This is the voluntary part of peripheral nervous system. and is also called autonomic nervous system (ANS). It comprises all those nerves, nerves fibres, ganglia, and plexus that carry impulses from CNS to viscera and from viscera to CNS. It regulates activity of smooth muscles, cardiac muscles and glands. Visceral or automatic system is further classified into sympathetic and parasympathetic systems.

1.3. Central Nervous System

1.3.1. Human Brain (Encephalon)

Location and Size: In central nervous system the anterior part is known as brain which is lodged in cranial cavity of the brain. The cranial bone protects the brain which weighs about 1.2-1.4 kg and it's made up of 10,000 million neurons. It is whitish, soft and flattened organ. The study of brain is known as encephalogy.

1.4. Division of Human Brain

The human brain is divided into three parts.

- Forebrain or Prosencephalon includes cerebrum, olfactory lobes and diencephalon.
- Midbrain or Mesencephalon includes crura cerebri and corpora quadrigemina
- Hindbrain or Rhombencephalon consists of Pons varolli, medulla oblongata, and cerebellum.[1]

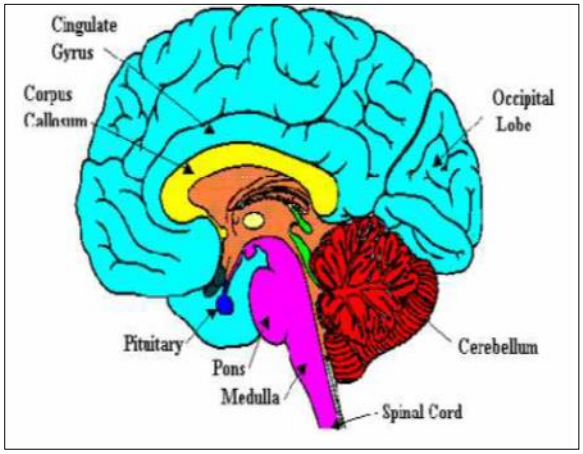

Figure 1 Saggital section

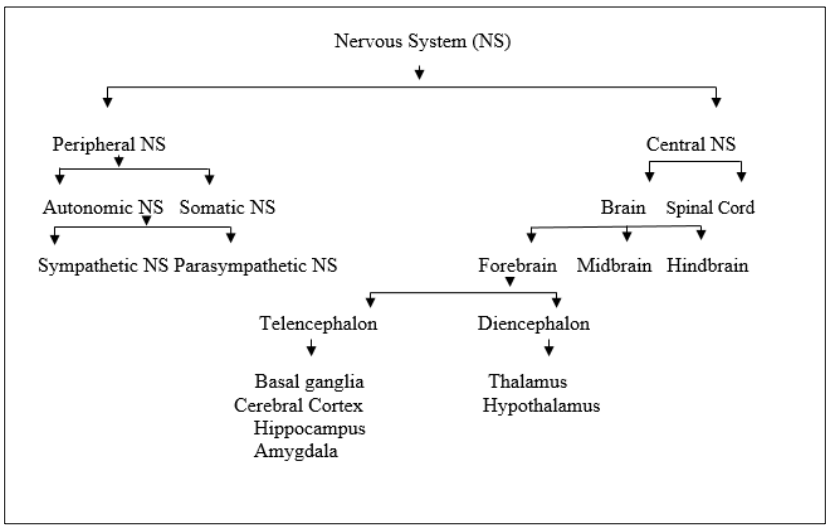

Figure 2 Classification of Nervous System

Neuronal death leads to neuronal degenerative disease which is one of the most common diseases all over the world. It is expected to increase nearly 131.5 million in 2050.

The reasons behind neuropharmacological diseases are due to genetic susceptibility factor, toxins like carbon monoxide, metamphetamin and heavy metal like ammonia.

The Mechanism is not known yet. [2]

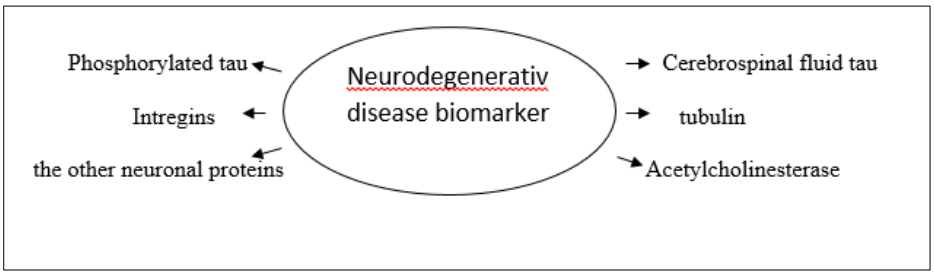

Figure 3 Biomarker for prediction of neurodegenerative diseases

1.5. Indian herbal market

The criteria for the selection of plants for herbal drug research for various human ailments are as follows:

- Medicinal plants uses in different part of the countries.
- Scientific literatures study on plants therapeutic efficacy.
- Naming the plant of those having therapeutic effect.
- Using plants having medicinal values for therapeutic purpose in countries as well as outside the country.

The first World Congress of Clinical Pharmacology and Therapeutics was held in London in 1980 to evaluate plant with therapeutic effect. Drug research on herbal consists of the following steps.

- Identifying plant which are used most often.
- Collecting of plant for research purpose.
- Taking plants to the research laboratory.
- Plant Storage
- For extract preparation
- For studying toxicity in animal with plant extract.
- To study therapeutic efficacy of the extract used in animal models.
- Extract Identification which is having more activities.
- Active molecule fractionation
- Structural elucidation of the bio-active molecule.
- T study the Bio-active molecule synthesis.[3]


Figure 4 Drug parts

- Scientific Name: *Moringa olifera*
- Family **:** Moringaceae
- Order **:** Brassicales
- Higher classification**:** *Moringa*
- Kingdom : Plantae
- Rank **:** Species
- Genus : *Moringa Adans*
- Species : *Moringa olifera* Lam. – horseradish tree, horseradish tree. [4]

Moringa olifera Lam., family Moringaceae, known as drumstick also called horse-radish tree, has a good nutritional source. These plant contain amino acids, phenolices, steroids, cyanates, flavanoids and alkaloids have been reported and also essential oil.

Health benefits of *M. olifera* are:

It is anti diabetic, anti inflammatory, helps to maintain healthy cholesterol level, can purify water and protect our heart.

Different parts of *M. olifera* have been shown to possess anti-microbial , anti-dyslipidemic, anthelmintic , helps in immunity boosting ,development in cardiovascular system , anti-cancer , anti-diabetic , anti-fatigue , antioxidant , analgesic, anti-inflammatory , and anti-toxic properties , as well as having effects on blood parameters , and a reproductive action.

A leaf extract of *M. olifera* and some of its constituents were also shown to have inhibitory effects on chemiluminescence and chemotactic activity of phagocytes.

M. olifera acts on the central nervous system (CNS) and have an anti-convulsant action. Such activity is due to its action on a central mechanism, which is affect in the release of γ-amino butyric acid (GABA).

Therefore, *Moringa olifera* has been used generally for the treatment of epilepsy. *M. olifera* has protective effects against degenerative as well as chronic neuronal diseases like Alzheimer's disease. It works on the neurons in the hippocampus to enhanced effects on memory. It also own an induction effect on myeloid cells differentiation and photoreceptors and to raise the development of neurons in the hippocampus.

Induced sleeping time prolongation is also induced by the plant by increasing the serum level of serotonin (5-HT), which help in the activation of the reticular-activating system that is needed for the sleep mechanism .The plant also induces a CNS depression in a dose-dependent manner in mice and sedative-hypnotic activity. [5]

1.6. Drug Profile

- **Diazepam**: - It belongs to benzodiazepam class commonly used for treatment on insomnia, anxiety, seizures including status epilepticus, muscle spasms, restless legs syndrome, alcohol withdrawal, benzodiazepine withdrawal and Ménière's disease.

- **MOA**: - it binds to a specific subunit on the GABA receptor at a site that is distinct from the binding site of the endogenous GABA molecule. The GABA receptor have an inhibitory channel which, decreases neuronal activity, when gets activated.
- **Paracetamol**: - Paracetamol, also known as acetaminophen, is a medication used to treat fever and mild to moderate pain. At a certain dose, paracetamol moderately decreases body temperature; it is inferior to ibuprofen, and the benefits of its use for fever are unclear.
- **MOA**: - Acetaminophen belongs to analgesics (pain relievers) class of drugs and antipyretics (fever reducers). The mechanism of action of acetaminophen is not yet known. It reduces the production of prostaglandins in the brain. Prostaglandins are chemicals that cause inflammation and swelling.
- **Pentazocin**: - Pentazocin injection is used to relieve pain from moderate to severe.
- **MOA**:-It is a potent analgesic belongs to narcotic analgesic group (pain medicine). It mostly acts on central nervous system.
- **Phenobarbitone Sodium**: It is a short acting barbiturate commonly known as sedative hypnotic drug.
- **MOA**:-Phenobarbiton increases synaptic inhibition acts on GABA receptors. It also inhibits calcium channel blockers. It is metabolized in liver and renally excreted. [6]

2. Materials and methods

2.1. Extraction of plant material

The whole *Moringa olifera* plant was collected in November 2022 from West Burdwan district, West Bengal (India). The plant specimen was authenticated from GCTS, Asansol (Voucher specimen no. YMA1) by Dr. Manik Baral.

Plant material was dried under shade and coarsely powdered for extraction. The coarsely powdered whole plant (150g) of *Moringa olifera* was subjected to extraction using methanol for 3 days by cold maceration. The methanolic extracts were concentrated by rotary vacuum evaporator under reduced pressure and then dried in open air.

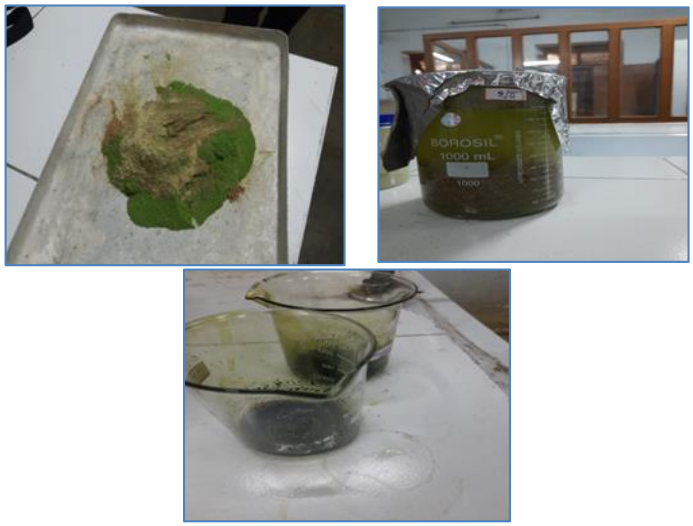

Figure 5 Extract

2.2. Animals

Wister albino mice which weigh about 30 grams were used for the study. The animals were kept in polypropylene cages. The mice were managed under standard laboratory conditions (250 + 2 0C; 12hr dark and light cycle). The animals were fed with standard diet and water. Ethical clearance (for handling of animals and the procedures used in study) was acquired from the Institutional Animal Ethical Committee before performing the study on animals [Protocol no.: GCTS/IEC/2021/AUG/09].

2.3. Pharmacological evaluation

2.3.1. Animal study

Male albino-Wistar mice weighing 30 g were used in the present study.

2.3.2. Housing and feeding condition:

All the mice were kept at room temperature (22±30˚C) in the animal house. The animals were treated and housed as per guidelines of Institutional Animal Ethics Committee (IAEC). Prior to the experiments, mice were fed with standard food and were acclimatized to standard laboratory conditions of temperature (22±30 ℃) and maintained an 12:12 hr light: dark cycle. They were provided with regular food and distilled water. All the experimental procedures were performed on animal after approval from the ethics committee and in accordance with the recommendations for the proper care and use of laboratory animals.

2.3.3. Extract:

Methanolic extract of *Moringa olifera* leaf

2.3.4. Standard drug used:

- Paracetamol (50 mg/kg)
- Diazepam (4 mg/kg, 2mg/kg & 1mg/kg)
- Pentobarbitone (40mg/kg)
- Pentazocin (50mg/kg)

2.4. Evaluation of Peripheral analgesic activity:

2.4.1. Writhing test

Peripheral analgesic activity for *M.Olifera* was evaluated using writhing test by Koster R et al., 1959.[17] Albino male mice (30-40 g) will be grouped into five groups of six animals each.

Table 1 Writhing test

Groups	Treatment
Group-I	control (received only vehicle)
Group-II	Standard drug paracetamol (50 mg/kg, i.p.)
Group-III	100 mg/kg b.w. methanolic extract
Group -IV	200 mg/kg b.w. methanolic extract
Group- V	400 mg/kg b.w. methanolic extract

Before administration of acetic acid, extracts and standard drug, each mice will be placed in the glass beakers. 0.1 ml of 0.6 % of acetic acid solution will be injected intraperitoneally to each animal. Then the animals will be allowed to elapse for 5 minutes, after administration of acetic acid and number of writhes will be recorded individually to each animal for the period of 30 minutes.

2.5. Evaluation of spontaneous locomotor activity: -

2.5.1. Locomotor activity

The healthy adult albino mice (30-40 g) will be firstly divided into five groups containing six animals each.

Table 2 Locomotor activity

Groups	Treatment
Group-I	control (received only vehicle)
Group-II	Standard drug diazepam (4 mg/kg i.p.)
Group-III	100 mg/kg b.w. methanolic extract
Group-IV	200 mg/kg b.w. methanolic extract
Group-V	400 mg/kg b.w. methanolic extract

After thirty minutes of standard drug and extract administration, each animal will be placed in photoactometer individually and then the locomotor activity will be counted for 10 minutes by Dewan S et al, 2000, Amos s et al 2001. [18,19]

2.6. Evaluation of muscle relaxant activity: -

2.6.1. Motor coordination (Muscle relaxant activity)

For the assessment of the motor co-ordination test, albino mice (30-40 g) will be divided into five groups containing six animals each group.

Table 3 Motor coordination

Groups	Treatment
Group-I	Control (received only vehicle)
Group-II	Standard drug diazepam (2 mg/kg, i.p.)
Group-III	100 mg/kg b.w. methanolic extract
Group-IV	200 mg/kg b.w. methanolic extract
Group-V	400 mg/kg b.w. methanolic extract

Rota-rod device will be used for the assessment of the experiment. At a rate of 16 revolutions per min the mice will be placed on the Rota rod. The fall off time for each animal will be noted. At the interval of 30 minutes for 3h the difference in the fall off time from the rotating rod will be noted by Ozturk et al., 1996; Perez et al., 1998. [20,21]

2.7. Evaluation of sedative activity: -

2.7.1. Effect on Phenobarbitone sodium sleep

For the evaluation of phenobarbitone sodium induced sleeping test, albino mice (30-40 g) will be divided into five groups containing six animals each group.

Table 4 Effect on Phenobarbitone sodium sleep

Groups	Treatment
Group-I	Control (received only vehicle)
Group-II	Standard drug diazepam (1 mg/kg, i.p.)
Group-III	100 mg/kg b.w. methanolic extract
Group-IV	200 mg/kg b.w. methanolic extract
Group-V	400 mg/kg b.w. methanolic extract

Phenobarbitone sodium (40 mg/kg b.w.) will be administered to individual animal later than 30 minutes of extract and standard drug administration. The onset of sleep and extent of sleep with condition being loss of righting reflex will be observed on the animals by Wambebe et al, 1985; Rolland et al., 1991. [22,23]

2.8. Experimental Procedure:-

2.8.1. Evaluation of Central analgesic activity:

Hot plate test

Central analgesic activity for *Moringa olifera* whole plant was evaluated using hot plate method as per described by Woolfe and MacDonald et al;1944.[24] Albino male mice will be divided into five groups containing six animals each.

Table 5 Hot plate test

Groups	Treatment
Group-I	**Control (received only vehicle)**
Group-II	Standard drug pentazocine (50 mg/kg, i.p.)
Group-III	100 mg/kg b.w. methanolic extract
Group- IV	200 mg/kg b.w. methanolic extract
Group -V	400 mg/kg b.w. methanolic extract

Individual mice will be placed on the hot plate which was maintained at 55 °C ± 1 ^{0}C, and latency of nociceptive response such as jumping, flicking of a hind limb and licking will be noted. And after administration of extracts, the readings will be recorded at 30, 60, 90 minutes time interval. The experiment will be terminated by 20 second after their placement on the hot plate to avoid damage to the paws of animals.

3. Results

Table 6 Analgesic activity of methanolic extract of *M. olifera* by acetic acid induced method

Group	Treatment	Number of Writhing
1	Control	60.83±2.91
2	Paracetamol(50mg/kg)	18.16±3.02
3	100 mg/kg b.w. methanolic extract	38.5±2.43
4	200 mg/kg b.w. methanolic extract	27.5±2.62
5	400 mg/kg b.w. methanolic extract	26.5±2.62

Results are expressed as ± SEM (n=6). Data processed by one way ANOVA,,*p<0.01 significant when compared to standard group. *p<0.01significant when compared to control group.

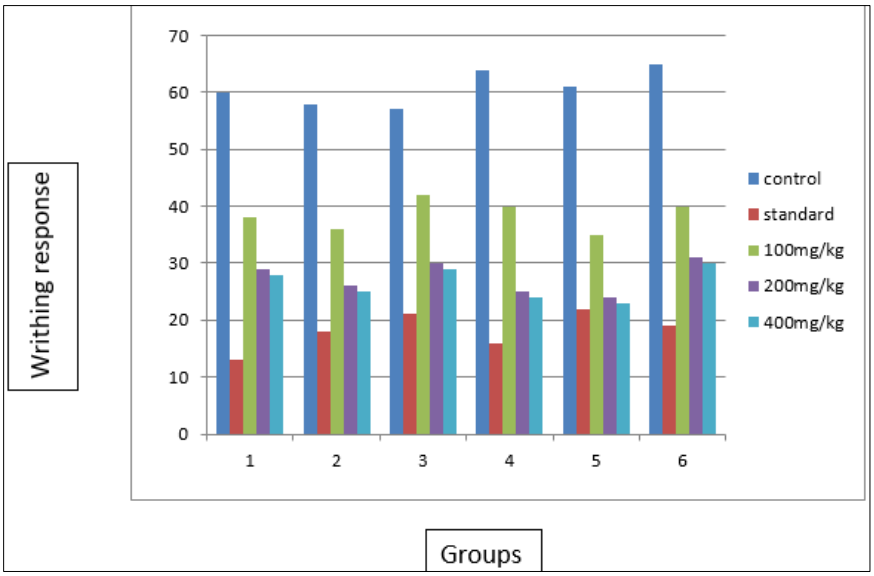

Figure 6 Graphical representation of acetic acid induced method.Y axis is writhing response, X is axis groups

Table 7 Spontaneous locomotor activity

Group	Treatment	Locomotor activity observed for 10 mins	
		Before dosing	After dosing
1	Control	170±0.35	170.82±0.30
2	Paracetamol(50mg/kg)	170±0.28	79.89±1.99
3	100 mg/kg b.w. methanolic extract	170±0.45	129.66±3.03
4	200 mg/kg b.w. methanolic extract	170±0.65	107.33±2.62
5	400 mg/kg b.w. methanolic extract	170±0.23	90.66±7.24

Results are expressed as ± SEM (n=6). Data processed by one way ANOVA p<0.01 significant when compared to standard group. *p<0.01 significant when compared to control group.

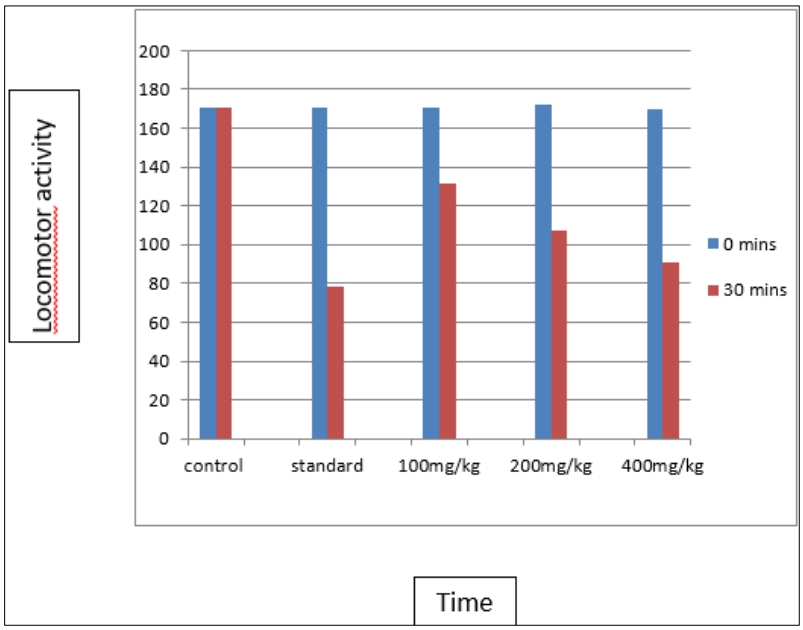

Figure 7 Graphical representation of spontaneous locomotor activity. Y axis is locomotor activity, X axis is time

Table 8 Muscle Relaxant Activity

Groups	Treatment	Fall of time in seconds (sec		
		30	60	120
1	Control	36.33±0.74	36.86±1.46	36.33±1.10
2	Paracetamol(50mg/kg)	25.66±0.74	22.5±1.89	15.5±2.36
3	100 mg/kg b.w. methanolic extract	35.16±0.68	33.16±0.68	31.33±1.10
4	200 mg/kg b.w. methanolic extract	32.5±0.95	30.66±1.37	25±1.06
5	400 mg/kg b.w. methanolic extract	30.5±0.95	25.5±0.95	23.33±1.69

Results are expressed as ± SEM (n=6). Data processed by one way ANOVA followed by Dunnett's test,*p<0.01 significant when compared to standard group. *p<0.01 significant when compared to control group.

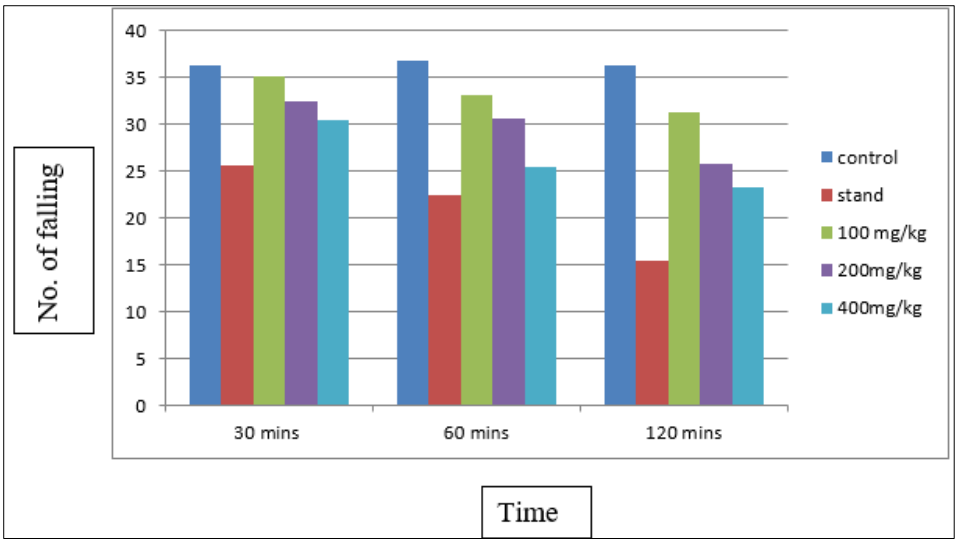

Figure 8 Graphical representation of muscle relaxant activity Y axisi is No. Of falling, X is axis time

Table 9 Phenobarbitone sodium sleep

Groups	Treatment	Mean sleeping time in mins.
1	Control	22±1.29
2	Paracetamol(50mg/kg)	69.5±2.62
3	100 mg/kg b.w. methanolic extract	30.33±1.88
4	200 mg/kg b.w. methanolic extract	37.66±2.62
5	400 mg/kg b.w. methanolic extract	48.16±1.57

Results are expressed as ± SEM (n=6). Data processed by one way ANOVA followed by Dunnett's test,*p<0.01 significant when compared to standard group. *p<0.01 significant when compared to control group.

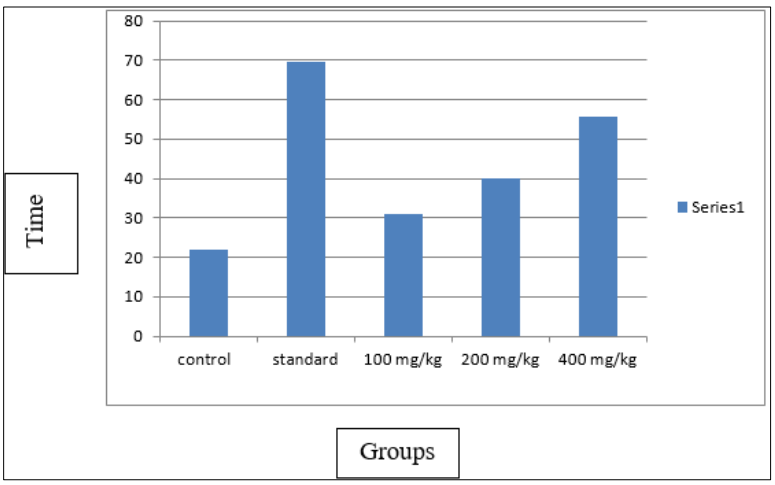

Figure 9 Graphical representation of phenobarbitone sodium sleep. Y axis is time, X axis is group

Table 10 Hot Plate Method

Groups	Treatment	Fall of time in seconds (sec)		
		30 mins	60 mins	120 mins
1	Control	4.66±0.74	5±0.81	4.83±0.89
2	Paracetamol(50mg/kg)	11±0.75	15.16±1.06	19.16±1.34
3	100 mg/kg b.w. methanolic extract	6.16±1.06	9.33±1.01	11.5±1.38
4	200 mg/kg b.w. methanolic extract	6.66±0.94	10.5±1.25	12.66±1.01
5	400 mg/kg b.w. methanolic extract	9.5±1.25	12.66±1.49	17.33±1.24

Results are expressed as ± SEM (n=6). Data processed by one way ANOVA followed by Dunnett's test,*p<0.01 significant when compared to standard group. *p<0.01 significant when compared to control group.

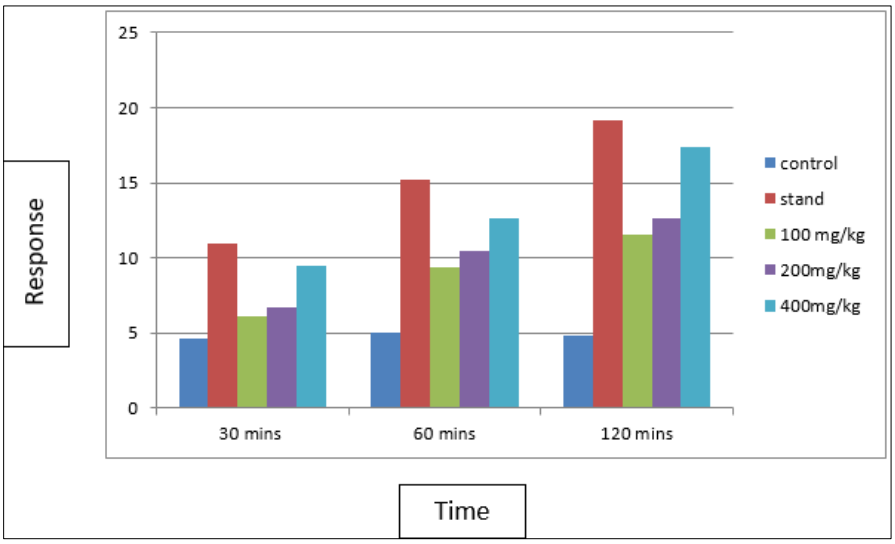

Figure 10 Graphical representation of hot plate method. Y axis is response , X axis time

4. Discussion

The present study involves the neuropharmacological study of ethanolic extract of *Moringa olifera* on behavioral studies such as Writhing effect, Actophotometer study, Potentiation of Phenobarbitone Sleeping Time, Rotarod Test in mice was performed.

These screening models are classic models for evaluating Neuropharmacological actions providing information about Spontaneous Locomotor activity, Peripheral analgesic activity, Central analgesic activity, Muscle Relaxant Activity, Sedative Activity.

The present study showed the effect of ethanolic extracts of *Moringa olifera* on Analgesic activity was evaluated by writhing test in mice. The result shown that the mean writhing effect produced in test group (200 mg/kg) is 27.5±2.62 showed less effect while test (400 mg/kg) group produced significant effect 26.5±2.62 compared to standard group 18.16±3.02. The results show that the test1, test2 concentrations of ethanolic extract of *Moringa olifera* produced a dose dependent writhing effect in experimental mice.

The Psychomotor activity of ethanolic extract of *Moringa olifera* was studied using Actophotometer activity cage. The locomotor activity score is taken as parameter for evaluation. The activity score was reduced in group 3 (100 mg/kg) from 170±0.45 to 129.66±3.03, group 4(200 mg/kg) from 170±0.65 to 107.33±2.62, group 5(400 mg/kg) 170.55±0.23 to 90.66±7.24. Thus *M.olifera* produced a dose dependent decrease in locomotor activity in mice.

The effect of ethanolic extracts of *Moringa olifera* on relaxant activity was evaluated by Rotarod method. The falling off time and number of fallings were parameter evaluated. There is decrease in time in falling in group 3(100 mg/kg) 31.55±1.10, group 4(200 mg/kg) 25±1.06 and group 5 (400 mg/kg) 24.5±1.70 and standard 21.5±3.09, before and after administration of the extract and standard compounds. These results shows ethanolic extracts of *M.olifera* produce a gradual decrease in no of falling & gradual decrease in falling off time in experimental test mice compared to standard while control group showed no significant variation.

The effect of ethanolic extracts of *Moringa olifera* on Sedative activity was evaluated by Potentiation of Pentobarbitone Sleeping time. Mean sleeping time was evaluated. The result showed that group 3(100 mg/kg) 30.33±1.88, group 4(200 mg/kg) 37.66±2.62 and group 5 (400 mg/kg) 48.16±1.57 and standard 69.5±1.57. This showed that ethanolic extracts of *M.Olifera* produced a dose dependent sedation in experimental mice.

The effect of ethanolic extracts of *Moringa olifera* on Central analgesic activity was evaluated by Hot Plate Method. The result showed mean score produced in group 3(100mg/kg) 6.16±1.06, 9.33±1.01, 11.5±1.38 showed less while group 4 (200mg/kg) 6.66±0.94, 10.5±1.25, 12.66±1.01 and standard 11±0.75, 15.16±1.06, 19.16±1.34. This showed that ethanolic extracts of *M.Olifera* produced a dose dependent central analgesic activity in experimental mice.

5. Conclusion

The present study shade dried leaves of *Moringa olifera* belonging to family Moringaceae having medicinally important active constituent is reviewed, with special emphasis on the biological activities. It has been studied to report on neuropharmacological activities.

Using many parameters such as the Writhing effect, Actophotometer study, Potention of Phenobarbitone Sleeping Time, Rotarod Test in mice, methanolic extract of *Moringa olifera* which at dosage (100,200&400mg/kg) exhibit a significant neuropharmacological effect. These findings demonstrate that the extract *M.olifera* is a mixture of many chemicals, and fractionation of the extract could results in neuropharmacological activity. Since the plant extract considerably reduced the number of writhes generated by acetic acid. Since no mice was found death so this herb is safe to use.

However, further investigation is underway to determine the exact reasons that are responsible for CNS depressant activity of ethanolic extract of *M.olifera* and the receptors involved for the execution of the activity.

Compliance with ethical standards

Acknowledgments

The authors are thankful to the Management and Principal, Gupta College Of Technological Sciences, Asansol for providing the facilities to carry out this study

Disclosure of conflict of interest

There is no conflict of Interest.

Statement of ethical approval

For performing experiment on animals, ethical clearance for the study was accorded by I.A.E.C. of Gupta College Of Technological Sciences, Asansol, (bearing registration number 955/PO/Re/S/06/CPCSEA) in resolution number GCTS/IEC/2021/Aug/09. The In vitro methods were executed by using Albino mice (male). Animals were obtained from the animal house of Gupta College Of Technological Sciences, Asansol.

References

[1] KNRN Publishers: Biology- Genetics By: Dr. Veer Bala Rastogi. Knrnpublications.com. Retrieved 19 April 2014.pg-38-45.

[2] Mahmoud M Elalfy. Protective Effect of Medicinal Plants on Neuron Degeneration Diseases and Controversial Predictive Diagnosis. Am J Biomed Sci & Res. 2019 - 2(4).AJBSR. MS.ID.000599. DOI: 10.34297/AJBSR.2019.02.000599

[3] Herbal Medicine: Current Status and the Future, 1) Sanjoy Kumar Pal, 2) Yogeshwer Shukla.

[4] Thripathi KD, medicinal pharmacology. Sixth edition, jayapeee brother medical publishers (p) ltd, kolkatha, 2008, 637-38.

[5] Al-Abria M, Ashiquea M, Ramkumara A, Nemmarb A and Badreldin H. Alia,. Motor and Behavioural Effects of Moringa oleifera Leaf Extract. NPC Natural Product Communications, 89

[6] Pezutto, J.M.: Biochem. Pharmacol. 53(2):121-133 (1997).

[7] Kumar R S, Sundram S, Sivakumar P, Nethaji R, V.Senthil, N. Venkateswara Murthy and Kanagasabi R CNS activity of the methanol extracts of Careya arborea in experimental animal model Bangladesh J Pharmacol 2008; 3: 36-43.

[8] Das S, Sasmal D, and BASU S. P Evaluation of CNS Depressant Activity of Different Plant parts of Nyctanthes arbortristis Linn. Indian J Pharmacol December 10, 2008, 36-43.

[9] Siddhuraju P , Becker K 2003 Apr "Antioxidant properties of various solvent extracts of total phenolic constituents from three different agro climatic origins of drumstick tree (Moringa oleifera Lam.) leaves".

[10] Ganguly R, Hazra R, Ray K, and Guha D, "Effect of Moringa oleifera in experimental model of Alzheimer's disease: role of antioxidants," Annals of Neurosciences, vol. 12, pp. 36–39, 2005.

[11] Umoh1 I.U, Edagha I.A and Aquaisua A.N 2018 "Neuroprotective Effect of Ethanol Extract of Moringa oleifera Leaf on the Neurofibres of Cerebellum of Quinine-treated Adult Wistar Rats".

[12] Sanchez M, Gonzalez E, Iglesias I, Lozano R and Gómez Serranillos PP-76- 9,2019.

[13] R. Ganguly, R. Hazra, K. Ray, and D. Guha, "Effect of Moringa oleifera in experimental model of Alzheimer's disease: role of antioxidants," Annals of Neurosciences, vol. 12, pp. 36–39, 2005.

[14] Stohs SJ , and Hartman MJ (2015), Review of the Safety and Efficacy of Moringa oleifera . Phytother. Res., 29, 796–804. doi: 10.1002/ptr.5325.

[15] Singh g. K. And kumar, v. Neuropharmacological screeningand lack ofanti depressantactivity of standardized extract of fumaria indica: apreclinical study Electronic Journal of Pharmacology and Therapy Vol. 3, 19-28 (2010).

[16] Ambavade S.D, Mhetre S.D, Tate V.D, Bodhankar S.L pharmacological evaluation of extract of Spearanthus indicus flowers on anxiolytic activity in mice Indian J Pharmacol | August 2006 | Vol 38 | Issue 4 |

[17] Koster R., Anderson M., De Beer E. J. Acetic acid for analgesic screening. Fed. Proc., 959; 18: 412.

[18] Dewan S., Sangraula H., Kumar V. L., Preliminary studies on the analgesic activity of latex of Calotropis procera. J. Ethnopharmacol., 2000; 73 (1-2): 307– 311.

[19] Amos S., Kolawole E., Akah P., Wambebe C., Gamaniel K., Behavioural effects of the aqueous extract of Guiera senegalensis in mice and rats. Phytomed, 2001; 8: 356–361.

[20] Ozturk Y., Aydine S., Baser K. H. C., Berberoglu H. Effects of Hypericum perforatum L. and Hypericum calycinum L. Extracts on the central nervous system in mice. Phytomed, 1996; 3: 139–146.

[21] Perez G. R. M., Perez L. J. A., Garcia D. L. M., Sossa M. H. Neuropharmacological activity of Solanum nigrum fruit. J. of Ethnopharmacol, 1998; 62: 43–48.

[22] Wambebe C. Influence of some agents that affect 5-HT metabolism and receptors and nitrazepam-induced sleep in mice. British J. of Pharmacol, 1985; 84:185–191.

[23] Rolland A., Fleurentain J., Lanhers M., Younos C., Misslin R., Morier F. Behavioural effects of American traditional plant Eschscholzia californica; sedative and anxiolytiproperties. Planta Medica, 1991; 57: 212–216.

[24] Woolfe G. and MacDonald A. D. The evaluation of the analgesic action of pethidine hydrochloride (Dermol). J. Pharmacol. Exp. Ther., 1944; 80: 300-330.