https://doi.org/10.48047/AFJBS.6.si2.2024.5833-5840

ANTIDEPRESSANT ACTIVITY OF AQUEOUS EXTRACT OF *EMBLICA OFFICINALIS* LEAVES IN ALBINO MICE

Vaibhavi Pandey[1*], Manoj Kumar Mishra[2], Mandvi Tiwari[3], Kashish Maurya[4]
[1234]Shambhunath Institute of Pharmacy, Prayagraj
Corresponding author: Vaibhavi Pandey; Email: thisisvaibhavi02@gmail.com

Volume 6 issue si2 2024

Received:15May2024

Accepted:10June2024

doi:10.48047/AFJBS.6. si2.2024.

5833-5840

ABSTRACT

In *EO*, the main constituent, i.e., tannic acid has non-selective monoamine oxidase activity. This study aims for assessing the EO leaves' potential as an antidepressant. The Forced swim test (F.S.T.) and the Tail suspending test (T.S.T.) in mice were the two models used in this work to evaluate the antidepressant activity in mice. After repeated oral therapy for 14 days of EO extract at rate of 100 & 200 mg/kg body weight significantly reduced the time of immobility. Additionally, leaves at this concurrent dose considerably down regulated the time of immobility in the TST following oral therapy respectively.

Keywords.: TST, FST, *Emblica officinalis*

INTRODUCTION

“Depressio," means "to sink". It is the Latin word that made the base for the origin of word Depression. Affective disorders such as depression are thought to be typified by mood swings, apathy, psychomotor slowness, and despair. It is believed that 8% of the general population suffers from depression. It is estimated that 121 million individuals worldwide suffer from depression.[1] Suicide is one of the most frequent consequences of depression, with near about 8.07% male and 12.06% female experiences a depression impact at some point in life.[2] Its signs and symptoms are very normal yet have a persistent difference from normal person.

Depression feels like a burden on the life on an individual and their relatives, which is generally characterized by a mood disfunction and lethargy. Ayurvedaa being the traditionally originated oldest system of medicines lists several plant-based single and compound medications that are used to address mental health issues. These compounds have been demonstrated to be as effective as their synthetic counterparts, while also having a less negative side effect profile.[3]

Emblica officinalis Gaertn, also referred to as Amla, is a minor genus of Emblica plants in the Euphorbiaceae family. Elagic acid, emblicannin A and B, punigluconnin, Galic acid and a few dozen flavonoids are among the chemical elements of *Emblica officinalis*, which are mostly tannins and compounds that resemble vitamin C in abundance. Ten percent gallic acid and thirty percent tannins are present in the EO's aqueous extract.[4]

MATERIALS AND METHODS

Plant Material

The *Emblica officinalis* leaves were gathered from a nearby Jhunsi, Prayagraj location. *Emblica officinalis* was confirmed to be authentic by the Botanical Survey of India (BSI). The Botanist there collected the *Emblica officinalis (Phyllanthus emblica* Linn.) leaves and verified the specimen. With voucher number SIP/2024/054, dated 14/03/2024, a specimen has been submitted with the BSI, Prayagraj. After the leaves were obtained, the adherent particles were separated from the plant substance, and the leaves were carefully cleaned. Following their mechanical crushing and shade drying, the leaves were powdered and subjected to various testing and analyses.

Reagents and Chemicals

The reference medication and the extract of test dosages were prepared using 1% gum acacia and saline as the carriers. The prescribed medication was imipramine (15 mg/kg).[5] Mice were given dexamethasone to induce depression-like behavior.[6]

Preparation of Extract of leaves of plant *Emblica officinalis*

Emblica officinalis leaves were powdered after being dried. For the aqueous extraction decoction method, the dried powder and distilled water are taken in 1:5 ratio and placed in a boiling water flask for an hour.[7] Filter the sol. with Whatmann filter paper no. 01 and keep in temperature between 4°C to 10°C for further use in experimental protocol.

Phytochemical Screening

The crude extract underwent phytochemical screening to look for secondary metabolites like glycosides, alkaloids, saponins, flavonoids, terpenes, and tannins.[8–10]

Experimental animals

The 25–30 gm albino mice were bought from Ms. Chakraborty Enterprises in Kolkata, West Bengal, India, and they were kept in Jhalwa, Prayagraj. They were kept at the temp. of 25 ± 3°C with a 12-hour light and 12 hours dark cycle. They have unrestricted access to water and were fed pellets. The study was conducted between 0900 and 1600 hours. The animal study protocol was verified by the I.A.E.C (Approval No. SIP/IAEC/003/03/24), dated March 18, 2024, and the animals were taken care for in accordance with CCSEA standards.

Acute toxicity Study

The OECD (Organisation for Economic Co-operation & Develapment) guideline no. 423 (Procedure for Acute toxic class technique) was followed in the performance of the acute oral toxicity investigation. Six groups of six healthy male albino mice each were randomly

assigned to groups. After an overnight fast with only water, the animals were given an intragastric tube containing aqueous extract of *Emblica officinalis* Gaertn. leaves at progressively higher dosing of 10, 100, 500 and 2000 milligram/kg in order to establish the acceptable dose. The animals were watched for general behavioral, neurological, and autonomic characteristics for one hour, then on a regular basis for four hours, and finally for a total of twenty-four hours.[11] Every half hour for the next twenty-four hours, and every day for the next 14 days, each mouse will be examined individually for signs of toxicity. Weight changes will be recorded twice a week, and mortality will be recorded every day. The LD50 of the leaf extract will be obtained by comparing the number of deaths with the predefined LD50 cut-off values in the guideline.[12]

Grouping and dosing of animals for screening of anxiety

Albino Swiss mice were used in this study and the groups were made as follows:

Group I: Vehicle administration group

Group II: Diseased control group

Group III: Standard (Imipramine, 15mg/kg)

Group IV: Test 01 (Aq. Extract of EO leaves 100mg/kg body weight)

Group V: Test 02 (Aq. Extract of EO leaves 200mg/kg body weight)

VARIOUS SCREENING MODELS FOR DEPRESSION

Tail suspension test

Our investigation followed the methodology outlined by Steru.[13] With the aid of adhesive tape, the animals were suspended by their tails 75 cm above the ground on a plastic rope. The time of immobility that was noted lasted for eight minutes. In the final 6 mins. of the behavioural study, the length of immobility was to be noted. Only when a mouse hung motionlessly and without movement was it deemed immobile.[14]

Forced swim test

In our investigation, the methodology outlined by Porsolt, et al. was applied. Every animal was kept separately in five-litre glass beakers that contain water to a level of fifteen centimetres, the animals were watched for six minutes.[15] The final four minutes of the observation period were used to record the length of immobility. When a mouse floated still or moved only when it was required to sustain the head on surface, it was said to be immobile. After every test, the water was replaced. [5]

Statistical analysis of study

This study data was analysed using GrafhPad Prism ver. 10, & its results were shown in mean ± standard error of mean (SEM). The mean difference was examined by One-way

analysis of variance (ANOVA). $p<0.05$ to $p<0.001$ was the range of level of statistical significance of the study.

RESULT

Phytochemical analysis of leaf extract

For extraction of phytochemicals implied in the study, two solvents were used for the extraction of the leaves of *EO*, distilled water and ethanol. The % yield of aqueous and ethanolic extract were:

Table 01: Percentage yield of leaf extract:

Extract	%Yield (w/w)
Distilled water	33.95%
Ethanol	27.35%

The aqueous extract of EO was subjected to preliminary evaluation of phytochemicals present in it. **Table 02** shows the presence of various phytoconstituents in Aq. leaf extract of EO.

Table 02: Phytoconstituents identification of leaf extract:

Sn. No.	Phytochemicals	Aq. Extract of leaf of *EO*
1	Alkoloid	+
2	Flavonoids	+
3	Carbohydrates	+
4	Tannins	+
5	Saponin	-
6	Steroid and triterpenoids	+
7	Glycosides	+
8	Phenols	+
9	Amino acids	+

Acute toxicity studies

Acute toxicity testing for EO was done on 150–180 gm body weight, healthy, male and female Swiss albino rats. Every animal was kept in room temperature polypropylene cages, fed

pellet food, and given access to water. Graded oral dosages of EO (5,50,300, and 2000 mg/kg) were administered. For the first six hours, the animals were watched continuously for gross effects, and after that, every six hours for the next 72 hours.[1] Animals responded favourably to dosages of up to 2000 mg/kg of emblica officinalis. At the prescribed dosages, EO was proven to be safe and no mortality was noted.[16]

Impact of EO Aqueous Extract on Immobile Durations in behavioural models:

When mice were given an aqueous extract (200.0 milligram/kg) for fourteen days in a row, the time of immobility in both behavioural models dramatically decreased, suggesting strong antidepressant-like effect.[17] However, the immobility times in TST alone were considerably reduced by the lower doze admin. of 100 milligram/kg. As a result, the aqueous extract had superior antidepressant-like effects. Also, the mice were given imipramine (15 milligram/kg) for 14 continuous days orally as the standard drug, the period of motionlessness in both the test models (Tail sus. test and For. swim test) were significantly decreased in comparison to the diseased group. This indicated a significant antidepressant-like effect in mice as shown in table below.[18] The statistical significance of the study was $P < 0.05$ to $P < 0.001$ when compared with control groups.

Table 03: Effects in Tail suspension test:

Group no.	Treatment for 14 days	Dose (/kg)	Immobility period (sec) [mean±SEM]
01	Vehicle (Distilled water)	10 ml	114 ± 0.32
02	Disease control group	-	209.3 ± 0.37
03	Imipramine	15mg	90.1 ± 0.54
04	Aq. Extract	100mg	133.5 ± 0.32
05	Aq. Extract	200mg	111.8 ± 0.39

Table 04: Effects in Forced swim test:

Group no.	Treatment for 14 days	Dose (/kg)	Immobility period (sec) [mean±SEM]
01	Vehicle (Distilled water)	10 ml	114.8 ± 0.36
02	Disease control group	-	185.6 ± 0.47
03	Imipramine	15mg	70.1 ± 0.60
04	Aq. Extract	100mg	137.1 ± 0.34
05	Aq. Extract	200mg	104.3 ± 0.27

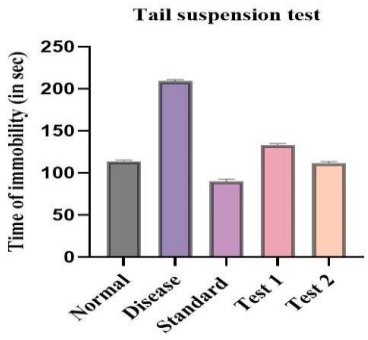

Figure 01: Outcomes of Imipramine and AEEO in the TST. The results (n = 6) are given as mean +/- S.E.M. whereas P < 0.001 to P<0.05 in rel. to the corresponding vehicle group.

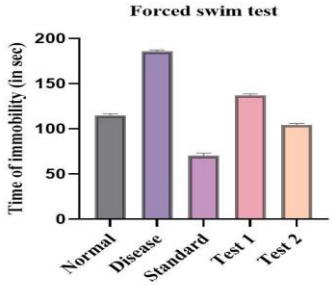

Figure 02: Outcomes of Imipramine and AEEO in the FST. The results (n = 6) are given as mean +/- S.E.M. whereas P < 0.001 to P<0.05 in rel. to the corresponding vehicle group.

DISCUSSION

Since only 50% of patients treated for depression with conventional antidepressants (TCAs, MOA inhibitors, SSRIs, and SNRIs) experience a complete recovery, there is growing curiosity in the investigation of the ability of combating depression of medicinal plants.[19] Potential antidepressants are frequently screened using the TST and FST. In both the Tail sus. test and For. swim test, antidepressants shorten the motionless period. It has been suggested that the immobile behaviour exhibited by mice during inevitable and unavoidable stressors is a reflection of behavioral desperation which may itself be a reflection of depressive illnesses in people.[20] In both models, there is a noteworthy relationship between the clinical potency and efficacy of antidepressants.[21]

E. officinalis was tested for its anti-depressant-like properties in animals using the Tail sus. test and For. swim test behavioral despair models, which are widely utilized. By calculating the shortened immobility duration that various antidepressant medications cause, these tests are frequently used in rats and mice to forecast anti-depressant ability.[2]

Here, mice received extract of leaves (200 milligram/kg) for 14 continuous days, and both the Tail sus. test and For. swim test showed a strong antidepressant effect. The immobility duration was shortened more by the 200 milligram/kg aq. extract than by the 100 milligram/kg lower dose. The highest potency of the extract was obtained at a dose of 200 milligram/kg since

smaller doses of the extract would not be sufficient for receptors.[22] Higher doses of the extract did not have any sedative effects, which may account for the notable reduction in immobility times as compared to the control.[23]

CONCLUSION

The results of this study have demonstrated the value of natural remedies in managing depression, and this complementary approach to care can provide a solid foundation for the creation of low-cost, secure, and potent medications.

REFERENCES

1. Badon H, Sharma P, Waheed SM, Singh S. Investigation of phytochemical composition, evaluation of antioxidant, antibacterial activities, and toxicity study of Emblica officinalis and Terminalia bellirica fruits. *Asian Journal of Pharmaceutical and Clinical Research*. 2016;9(6):96-102. doi:10.22159/ajpcr.2016.v9i6.13731
2. Pemminati S, Sahu S. *Antidepressant Activity of Aqueous Extract of Fruits of Emblica Officinalis in Mice*.; 2010. https://www.researchgate.net/publication/282860385
3. Sembulingam K, Namasivayam' A. *EFFECT OF OCIMUM SANCTUM LINN ON NOISE INDUCED CHANGES IN PLASMA CORTICOSTERONE LEVEL*. Vol 41.; 1997.
4. Variya BC, Bakrania AK, Patel SS. Emblica officinalis (Amla): A review for its phytochemistry, ethnomedicinal uses and medicinal potentials with respect to molecular mechanisms. *Pharmacol Res*. 2016;111:180-200. doi:10.1016/j.phrs.2016.06.013
5. Poleszak E, Wlaÿ P, Kêdzierska E, et al. Immobility stress induces depression-like behavior in the forced swim test in mice: effect of magnesium and imipramine. Published online 2006.
6. Peeters BWMM, Broekkamp CLE. *Involvement of Corticosteroids in the Processing of Stressful Life-Events. A Possible Implication for the Development of Depression*. Vol 49.; 1994.
7. Raja AM, Banji D. Evaluation of standardisation parameters, pharmacognostic study, preliminary phytochemical screening and in vitro antidiabetic activity of Emblica officinalis fruits as per WHO guidelines. *~ 21 ~ Journal of Pharmacognosy and Phytochemistry*. 2014;3(4):21-28.
8. Shaikh JR, Patil M. Qualitative tests for preliminary phytochemical screening: An overview. *Int J Chem Stud*. 2020;8(2):603-608. doi:10.22271/chemi.2020.v8.i2i.8834
9. Javale P, Sabnis S. *Antimicrobial Properties and Phytochemical Analysis of Emblica Officinalis*. https://www.researchgate.net/publication/228514313
10. Manke MB. PHYTOCHEMICAL ANALYSIS OF METHANOLIC EXTRACT OF EMBLICA OFFICINALIS LEAVES *Corresponding Author. Published online 2018. doi:10.20959/wjpps201811-12493
11. Nain P, Saini V, Sharma S, Nain J. Antidiabetic and antioxidant potential of Emblica officinalis Gaertn. leaves extract in streptozotocin-induced type-2 diabetes mellitus (T2DM) rats. *J Ethnopharmacol*. 2012;142(1):65-71. doi:10.1016/j.jep.2012.04.014

12. Detke MJ, Johnson J, Lucki I. *Acute and Chronic Antidepressant Drug Treatment in the Rat Forced Swimming Test Model of Depression*. Vol 5.; 1997.

13. Steru L, Chermat R, Thierry B, Simon P. The tail suspension test: A new method for screening antidepressants in mice. *Psychopharmacology (Berl)*. 1985;85(3):367-370. doi:10.1007/BF00428203

14. Cryan JF, Mombereau C, Vassout A. The tail suspension test as a model for assessing antidepressant activity: Review of pharmacological and genetic studies in mice. *Neurosci Biobehav Rev*. 2005;29(4-5):571-625. doi:10.1016/j.neubiorev.2005.03.009

15. Unal G, Canbeyli R. Psychomotor retardation in depression: A critical measure of the forced swim test. *Behavioural Brain Research*. 2019;372. doi:10.1016/j.bbr.2019.112047

16. *OECD/OCDE 423 OECD GUIDELINE FOR TESTING OF CHEMICALS Acute Oral Toxicity-Acute Toxic Class Method INTRODUCTION*.; 2001.

17. David DJP, Renard CE, Jolliet P, Hascoët M, Bourin M. Antidepressant-like effects in various mice strains in the forced swimming test. *Psychopharmacology (Berl)*. 2003;166(4):373-382. doi:10.1007/s00213-002-1335-4

18. Dhingra D, Joshi P, Gupta A, Chhillar R. Possible Involvement of Monoaminergic Neurotransmission in Antidepressant-like activity of Emblica officinalis Fruits in Mice. *CNS Neurosci Ther*. 2012;18(5):419-425. doi:10.1111/j.1755-5949.2011.00256.x

19. Nestler EJ, Barrot M, Dileone RJ, Eisch AJ, Gold SJ, Monteggia LM. *Review Neurobiology of Depression the Focus of Efforts to Understand the Pathophysiology*. Vol 34.; 2002.

20. Can A, Dao DT, Terrillion CE, Piantadosi SC, Bhat S, Gould TD. The tail suspension test. *Journal of Visualized Experiments*. 2011;(58). doi:10.3791/3769

21. Castagné V, Moser P, Roux S, Porsolt RD. Rodent Models of Depression: Forced Swim and Tail Suspension Behavioral Despair Tests in Rats and Mice. *Curr Protoc Pharmacol*. 2010;49(1). doi:10.1002/0471141755.ph0508s49

22. Wankhar D, Sheela Devi R, Ashok I. Emblica officinalis outcome on noise stress induced behavioural changes in wistar albino rats. *Biomedicine and Preventive Nutrition*. 2014;4(2):219-224. doi:10.1016/j.bionut.2013.12.011

23. Nagapati Prabhakar Bhat by. *EVALUATION OF ANTIDEPRESSANT LIKE ACTIVITY OF AQUEOUS EXTRACT OF FRUITS OF EMBLICA OFFICINALIS (AMLA) IN ALBINO MICE*.