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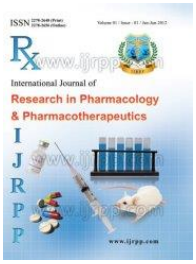

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An experimental study on antidepressant activity of methanolic extract of momordica charantia leaves

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ABSTRACT

Background

Momordica charantia commonly known as Karela is widely known for its medicinal properties. Its Antidepressant activity is least evaluated. Depression being common psychiatric illness of all age groups. Currently available Antidepressants have their own limitations. The antidepressant activity of Momordica Charantia leaves was evaluated in this study.

Aim

The aim of this study is to evaluate the Antidepressant activity of Methanolic extract of Momordica Charantia leaves.

Material and Methods

This study was done in Department of Pharmacology, JNMC, AMU. Antidepressant activity of MEMC (Methanolic extract of Momordica Charantia leaves) . at doses of 100mg/kg, 200 mg/kg and 300 mg/kg was evaluated in Swiss Albino mice and Albino Wistar rats in Tail suspension test, Forced swim test, Spontaneous motor activity, Learned helplessness test and 5-Hydroxytryptophan induced Head Potentiation tests.

Results

MEMC exhibited antidepressant activity by significantly decreasing the immobility time in Tail Suspension test in all doses and in doses of 200 mg/kg and 300 mg/kg in Forced swim test. No significant increase in Spontaneous motor activity was seen in all three doses ruling out psychostimulant activity. In Learned Helplessness test number of Escape failures was decreased at doses of MEMC 200 mg/kg and 300 mg/kg. Increase in Head twitches was seen in both MEMC 200 mg/kg and 300 mg/kg in 5-Hydroxytrytophan induced Head Potentiation in mice.

Conclusion

Antidepressant activity was exhibited by Methanolic extract of Momordica Charantia leaves in animal models of Depression.

Keywords: Depression, Momordica charantia, Tail suspension test, Forced swim test, Learned Helplessness test, 5-Hydroxytrytophan induced Head Potentiation in mice.

INTRODUCTION

Momordica charantia commonly known as Karela in Hindi is well known herb for its medicinal properties. Momordica charantia fruit is widely used in Indian cuisine. Antidiabetic [1], Antioxidant [2], Hepatoprotective [3], Nephroprotective [4], Antimicrobial [5] and Antiviral activity [6] have been demonstrated on leaves of Momordica charantia. Antidepressant activity of Momordica charantia leaves was least evaluated. [7]

Depression is the fourth leading cause of morbidity and economic loss, contributing to increase in mortality worldwide. Depression will be the second leading cause of disability burden, by the year 2020 as predicted by World Health Organization [8]. Depression accounts for 4.5% of the worldwide total burden of disease in terms of disability-adjusted life years. [9]

Depression affects all age groups. Depressed individuals of older age group also likely to suffer from comorbid conditions like Angina, Myocardial infarction, Diabetes mellitus, Cancer, Parkinson's disease and Alzheimer's disease. A good number (20%) of all depressed patients are refractory to the available antidepressants at adequate doses. It is compounded with a "therapeutic lag" lasting 3-4 weeks before therapeutic response becomes evident in them. [10] Antidepressant drugs are associated with many adverse effects [11] and interactions with the drugs prescribed for the treatment of comorbid conditions. [12]

Literature search revealed a single study on antidepressant activity of Momordica charantia leaves. Therefore this study was conducted to evaluate antidepressant activity of Momordica charantia leaves.

MATERIAL AND METHODS

Momordica charantia leaves were collected from fields of Pannipur village, Aligarh district, Uttar Pradesh. They were identified and authenticated by Prof. S. H. Afaq, Department of Pharmacognosy, Ilmul Advia, A.K. Tibbya College, Aligarh Muslim University, Aligarh. Specimen was deposited and voucher number (SC-0135/12) was obtained. Leaves were finely powdered in a grinder after they were dried. Powder was extracted in Methanol using Soxhlet's apparatus.

The study protocol was approved by the Institutional Animal Ethics Committee (IAEC) on 13.04.2012. All animal experiments were carried out as per the rules and regulations of IAEC & CPCSEA (Committee for the Purpose of Control and Supervision of Experiments on Animals) under the "Guidelines for Care and Use of Animals in Scientific Research".

ANIMALS USED

1. Wistar Albino rats of either sex (150-200 gm).
2. Swiss Albino mice of either sex (20-40 gm).

The animals were procured from the Central Animal House, JNMC, Aligarh Muslim University. They were housed in polypropylene cages bedded with paper strips under standard conditions and fed with pellet diet (Ashirwad Industries) and water ad libitum in the Pharmacology section of Central Animal House. They were acclimatized to laboratory conditions for 1 week before experimental study. No acute toxicity of Methanolic extract was reported even at the dose of 5000 mg/kg. [13] Dose was selected on the basis of previous studies.

GROUPING OF ANIMALS: Experimental Design

The animals were divided into 5 groups, consisting of normal control group, positive control group and 3 test groups. Each group consisted of 6 animals of either sex (n=6). Fresh animals were taken for each group in each screening method.

Table 1. Experimental design for each screening method.

Groups	Name	Treatment received
I.	Normal Control	Propylene glycol 1ml/kg, Orally.
II.	Positive Control	Imipramine/Fluoxetine*
III.	Test group- 1	Methanolic extract 100mg/kg, Orally.
IV.	Test group-2	Methanolic extract 200mg/kg, Orally.
V.	Test group-3	Methanolic extract 300mg/kg, Orally.

*Imipramine 15 mg/kg was used in all screening tests except 5-Hydroxytryptophan potentiation in mice in which Fluoxetine 20mg/kg was used.

SCREENING OF ANTIDEPRESSANT ACTIVITY

Tail suspension test [14]

Mice were treated with Drug/ Vehicle/ Methanolic extracts for 7 days. On 7th day test was carried out, 1 hour after Drug/ Vehicle/ Methanolic extract was administered. Mice were suspended by woollen thread secured with adhesive tape placed approximately 1 cm from the tip of the tail for 6 mins. Intermittent periods of immobility were recorded and there sum showed the total period of immobility. Mice were considered immobile when they hung passively and completely motionless.

Forced swim test [15]

Rats were administered with Drug/ Vehicle/ Methanolic extracts for 8 days. Rats were individually forced to swim inside a vertical Plexiglass cylinder (height: 28 cm; diameter: 20 cm, containing 20 cm of water maintained at 25 °C). On 7th day of drug administration, Pre test session was conducted for 15 mins. Test was carried out on 8^{th} day 1 hr after Drug/ Vehicle/ Methanolic extracts administration for 5 min. An animal was judged to be immobile whenever it remained floating passively in water (nose above water surface). Duration of immobility was recorded.

Spontaneous motor activity [16]

Rats were treated with Drug/ Vehicle/ Methanolic extracts for 7 days. Rats were acclimatized with Actophotometer before experiment. Spontaneous motor activity test was done on day 0(pre drug) and day 7(60 mins after drug administration on 7th day). They were placed in Actophotometer and the light beam interruptions were recorded as activity counts. Activity counts were assessed for a duration of 10 mins. Results of day 7 were compared with that of day 0.

Learned helplessness test [17]

Rats were administered with Drug/ Vehicle/ Methanolic extracts for 7 days. Rats were acclimatized in the conditional apparatus before experiment on 5^{th} and 7^{th} day of drug administration. On 5^{th} day rats were exposed to electric shock (30 V) on a schedule of 10s of shock/min for 1 h. Gate was closed during this period. On 7^{th} day, the gate was kept open and a 30 V shock initiated. Shock was terminated in 10s if rats didn't escape to the other side through the gate. If rat escapes, it was allowed to remain on the other side for the duration of 10s, then returned to the same chamber. Ten such trials were conducted with an intertrial interval of 20s. Number of escape failures was noted.

5-Hydroxytryptophan potentiation in mice [18]

Mice were treated with Drug/ Vehicle/ Methanolic extracts for 7 days. On 7^{th} day, 5-Hydroxytryptophan was given at a dose of 200 mg/kg i.p. after 1 hour of drug administration. After 15 mins, characteristic symptom of head-twitches was observed intermittently at 2 min intervals (19-21, 23-25, 27-29 mins) for 6 mins duration.

STATISTICAL ANALYSIS

Values were expressed as Mean ± SEM. Statistical significance was calculated by paired Student's t test (Spontaneous motor activity) and one way ANOVA followed by post hoc Dunnett's multiple comparison test using SPSS-17 software. $P<0.05$ was considered to be statistically significant.

RESULTS

Plant extracts

Green coloured semi solid mass of oily consistency with 7.85% yield was obtained by methanolic extraction.

Tail suspension test

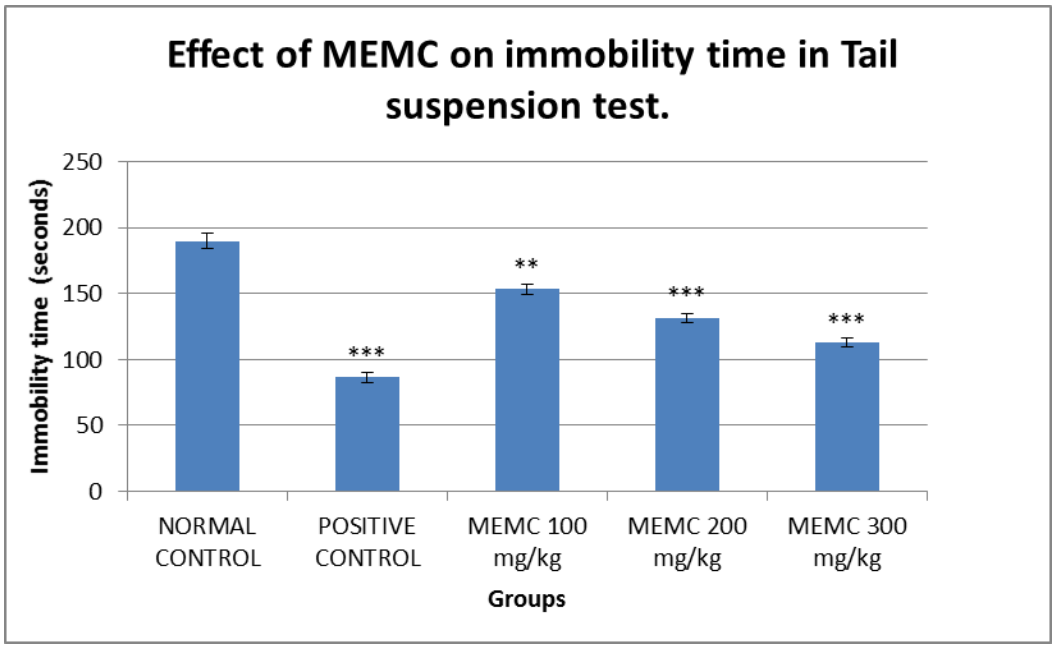

Fig 1. Effect of MEMC and Control drugs on Immobility time in Tail suspension test

MEMC: Methanolic extract of Momordica charantia leaves, n = 6 rats in each group.*- $p<0.05$, ** - $p<0.01$, *** - $p<0.001$. Level of significance compared to normal control group.Values are expressed as mean ± SEM.

Immobility time was significantly decreased ($p<0.001$) in positive control group compared to normal control group. Significant decrease in Immobility time was noted in MEMC 100 mg/kg ($p<0.01$), MEMC 200 mg/kg ($p<0.001$) and MEMC 300 mg/kg ($p<0.001$) received groups.

Forced swim test

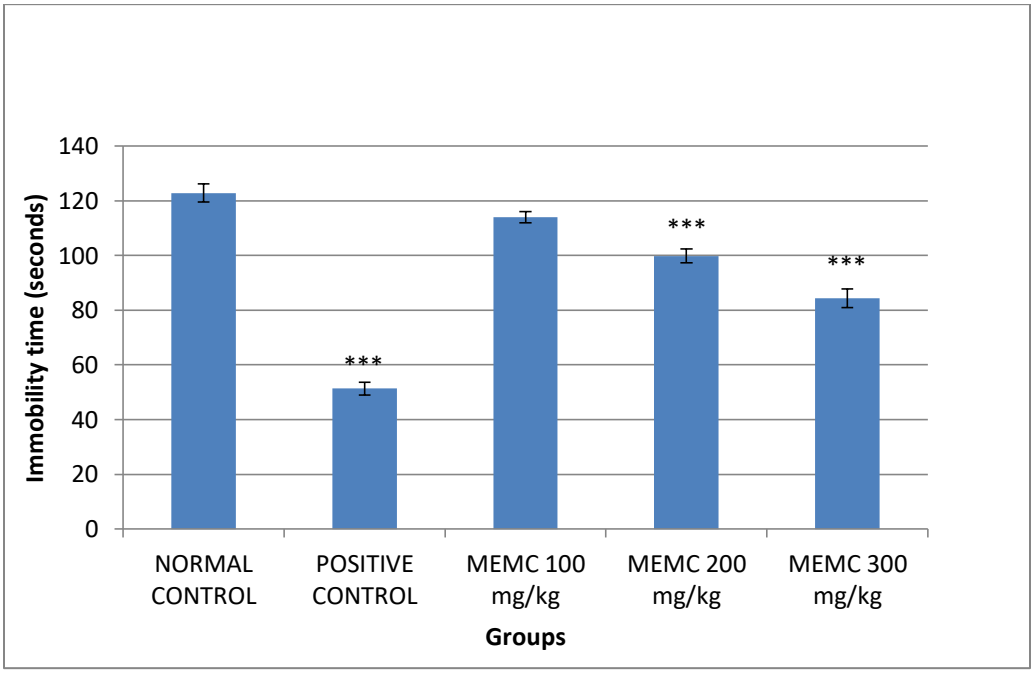

Fig 2. Effect of MEMC and Control drugs on Immobility time in Forced swim test.

MEMC: Methanolic extract of *Momordica charantia* leaves, n = 6 rats in each group.*- p<0.05, ** - p <0.01, *** - p <0.001. Level of significance compared to normal control group.Values are expressed as mean ± SEM.

Immobility time was significantly decreased (p<0.001) in positive control group compared to normal control group. No significant decrease in Immobility time was noted in MEMC 100 mg/kg received group. Significant decrease in Immobility time was noted in MEMC 200 mg/kg (p<0.001) and MEMC 300 mg/kg (p<0.001) received groups.

Spontaneous Motor Activity

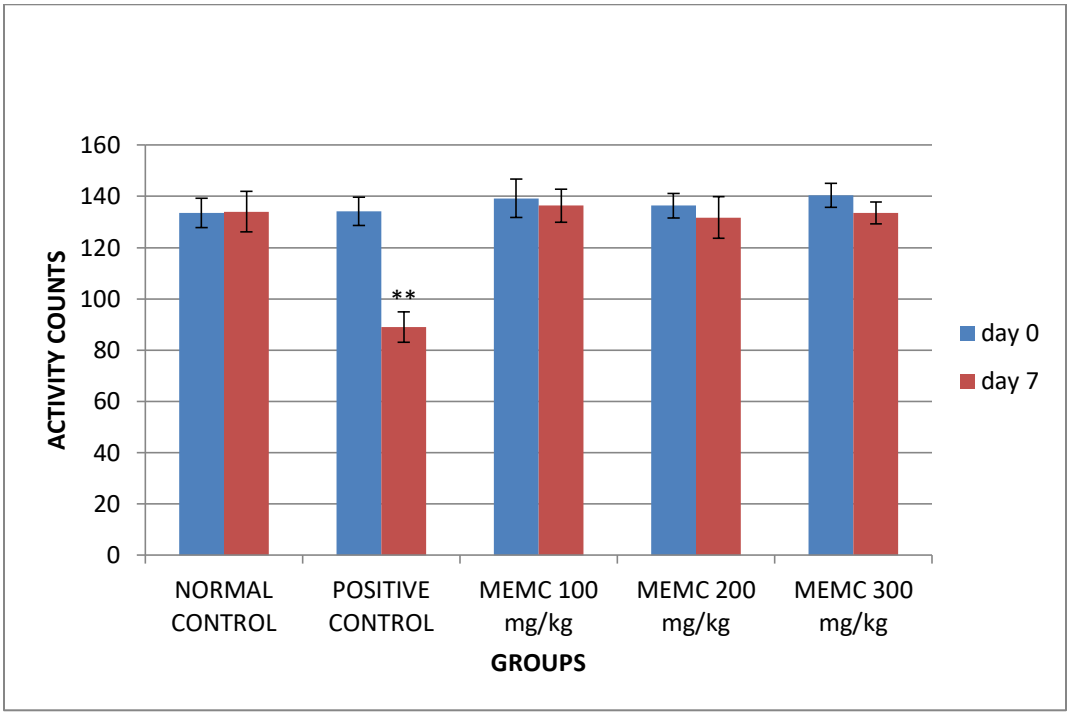

Fig 3. Effect of MEMC and Control drugs on Activity counts on day 0 and day 7.

MEMC: Methanolic extract of ***Momordica charantia*** leaves, n = 6 in each group*- p<0.05, **- p <0.01. Level of significance compared to normal control group. Values are expressed as mean ± SEM.

In normal control group there was no significant change in Spontaneous motor activity on day 7 compared to day 0, while in Imipramine 15 mg/kg treated group Spontaneous motor activity was significantly decreased (p<0.01) on day 7. This is due to its sedative effect.[19] No significant change in Spontaneous motor activity was seen with methanolic extract treated groups.

Learned Helplessness test

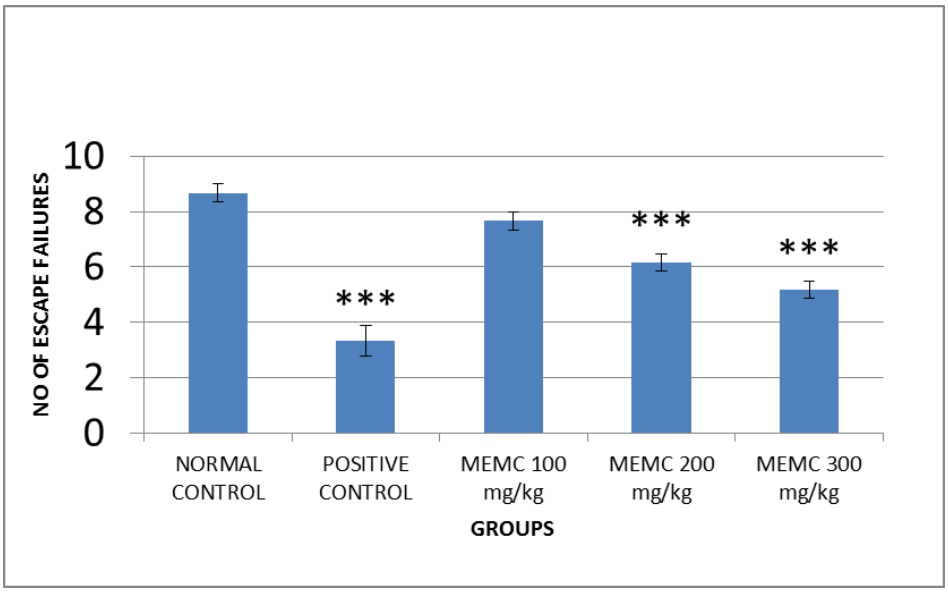

Fig 4. Effect of MEMC and Control drugs on Escape failures in Learned Helplessness test.

MEMC: Methanolic extract of *Momordica charantia* leaves, n = 6 rats in each group. *- p<0.05, ** - p <0.01, *** - p <0.001. Level of significance compared to normal control group.Values are expressed as mean ± SEM.

Significant decrease in Escape failures was seen in positive control (p<0.001) group. No significant decrease in Escape failures was seen in MEMC 100 mg/kg received group. Significant decrease in Escape failures was seen in MEMC 200 mg/kg (p<0.001) and MEMC 300 mg/kg (p<0.001) received groups.

5-Hydroxytryptophan potentiation in mice

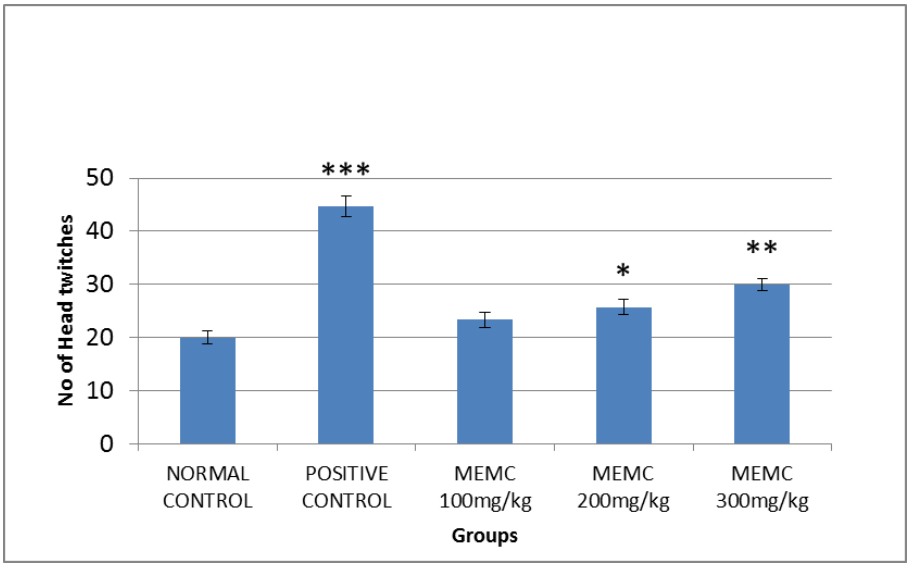

Fig 5. Effect of MEMC and Control drugs on Head twitches in 5-Hydroxytryptophan potentiation in mice.

MEMC: Methanolic extract of Momordica charantia leaves, n = 6 mice in each group.*- p<0.05, ** - p <0.01, *** - p <0.001. Level of significance compared to normal control group. Values are expressed as mean ± SEM.

Head twitches was significantly increased in Fluoxetine 20 mg/kg (p<0.001) compared to normal

control group. No significant increase in Head twitches was seen in MEMC 100 mg/kg received group. Significant increase in Head twitches was seen in MEMC 200 mg/kg($p<0.05$)and MEMC 300 mg/kg ($p<0.01$) received groups.

DISCUSSION

Depression is a heterogenous clinical disorder with varied aetiology. Depressive symptoms are subjective and varies from individual to individual. Animal models of depression does not resemble human depression pathologically. Instead the symptomatic profile of humans are simulated in animals. [20] Tail suspension test and Forced swim test are behaviour despair tests.

Tail suspension test is based on the hypothesis that when an animal is subjected to an untoward situation, animal either goes into state of "agitation" or state of "immobility". Antidepressant drugs shifts the balance towards "agitation". Mice are suspended by tail and duration of immobility is noted. [14]

Forced swim test is based on the hypothesis that when an animal is forced to swim in a restricted space, after sometime it goes into a state of immobility. This state of immobility is a state of despair in rats. Antidepressant drugs decrease the duration of immobility. [15]

Methanolic extract decreased immobility time in both the tests. Tail suspension test is more sensitive test compared to Forced swim test. [14] This may be the reason why the lower dose (100 mg/kg) is showing significant results in Tail suspension test and non significant results in Forced swim test.

Apart from antidepressants, Psychostimulants also decrease the duration of immobility in Tail suspension test and Forced swim test. [14, 15] In order to rule out the Psychostimulant effect of the extract (MEMC), its effect on Spontaneous motor activity in normal rats is seen. Antidepressants do not show any significant change in Spontaneous motor activity, whereas Psychostimulants increases it in normal rats. [14, 15]

Methanolic extract at all three doses did not show any significant change in Spontaneous motor activity, thereby its Psychostimulant effect was ruled out.

Learned Helplessness test was used to assess the antidepressant activity of Momordica charantia leaves. The antidepressant activity was evaluated by observing the number of escape failures of rats.

Animals are subjected to electric shock without a chance to escape, later on when the rats are subjected to same strength of electric shock with a chance to avoid it, they fail to do it. Antidepressants overcome this "Helpless" situation. [21]

Rats were subjected to electric shock of 30 volts on day 5, in a space in which there was no scope for escape. On 7th day, rats were subjected to same strength of shock and their ability to overcome the "Helpless" situation by escaping through the open gate was observed. Decrease in the number of escape failures means rats overcame the state of "Helpless" situation. Drugs with Antidepressant activity decrease the number of escape failures.

Methanolic extract in two doses 200 mg/kg ($p<0.001$) and 300 mg/kg ($p<0.001$) showed significant decrease in Escape failures.

Antidepressants increasing serotonergic neurotransmission increase the potentiation of 5-HTP in mice. [18] Increase in serotonergic transmission inhibits inhibitory neurons in mice and rats causes development of Hallucinogenic effect which presents as Head twitches (wet dog shakes). [22] Methanolic extract at doses of 200 mg/kg and 300 mg/kg showed significant effect on 5-HT potentiation implying that it is acting by increasing 5-HT levels in brain. This may be one of the mechanism of action for its antidepressant activity but not exclusive as screening tests for other mechanisms of action have not been carried out.

Methanolic extract 200 mg/kg and 300 mg/kg showed significant effect in all the screening models tested in the study. Methanolic extract 100 mg/kg showed significant effect in Tail suspension test but did not show significant effect in other screening methods. This may be due to the fact that Tail suspension test is more sensitive test than other screening tests. [14]

Methanolic extract of Momordica charantia exhibited antidepressant activity in this study. This activity can be attributed to the phytochemicals present in the methanolic extract of Momordica charantia. Tannic acid, Gallic acid, Catechin and other phenolic compounds are present in the leaves of Momordica charantia. [2] Tannic acid (30%) and Gallic acid (10%) present in Emblica officinalis were contributing for antidepressant activity evaluated by Tail suspension test and Forced swim test. Non-specific MAO inhibition was proposed to be the

mechanism of action of Tannic acid for its antidepressant activity. [23]

Gallic acid exhibited antidepressant activity by inhibiting MAO-A enzyme, thereby increasing the levels of monoamines in the synapse, which was evaluated by Forced swim test in mice. [24]

Catechin rich Butanolic fraction exhibited antidepressant activity in Forced swim test. Its action on Hippocampal synaptosomes showed decreased uptake of serotonin, Noradrenaline and Dopamine, thereby increasing the monoamine levels in the hippocampus. [25]

Depression is also attributed due to deficiency of Zinc. [26] Zinc supplementation is effective in overcoming the treatment resistant depression combined with antidepressants. [27] Concentration of Zinc in leaves is reported to be 350mg/kg of dry weight of leaves. [5] This may be contributing the action of Methanolic extract in animal models. However this study cannot conclude that it is the main mechanism of action as none of the screening models is known to decrease the concentration of zinc.

Tryptophan is present in Momordica charantia leaves. [28] The antidepressant effect of the extracts seen in the study may be due to Tryptophan which is the precursor of serotonin present in the leaves.

Momordica charantia leaves contain phenolic compounds Naringin and Rutin. [29] Naringin acting through nitric oxide mechanism has shown to be effective in post stroke depression by preventing the oxidative damage.[30] Rutin also exhibits antidepressant activity. [31]

In Momordica charantia, leaves show highest antioxidant power as shown by DPPHl(1,1-diphenyl-2-picrylhydrazyl free radical) radical-scavenging activity, Hydroxyl radical-scavenging activity, β-Carotene–linoleate bleaching assay, Ferric reducing/antioxidant power (FRAP) assay. Alkaloids, Flavonoids, Tannic acid and Gallic acid also known to exhibit antioxidant activity. This antioxidant activity may be countering the oxidative stress leading to depression. [2]

From this study it can be concluded that Antidepressant activity was exhibited by Methanolic extract of Momordica charantia leaves, probably by increasing 5-HT levels in brain. Further studies are needed to confirm the same.

REFERENCES

[1]. Tarkang PA, Ofogba CJ. Evaluation of the Hypoglycemic Activity and Safety of Momordica charantia (Cucurbitaceae). African Journal of Pharmaceutical Sciences and Pharmacy, 3(1), 2012, 17-29.

[2]. Kubola J, Siriamornpun S. Phenolic contents and antioxidant activities of bitter gourd (Momordica charantia L.) leaf, stem and fruit fraction extracts in vitro. Food Chemistry. 110, 2008, 881–890.

[3]. Chaudhari BP, Chaware VJ, Joshi YR, Biyani KR. Hepatoprotective activity of Hydroalcoholic extract of Momordica charantia Linn. leaves against Carbon tetrachloride induced Hepatopathy in Rats. International Journal of ChemTech Research. 1(2), 2009, 355-358.

[4]. Chaware VJ, Chaudhari BP, Joshi YR, Biyani KR. Protective Effect of the Aqueous Extract of Momordica charantia Leaves on Gentamicin Induced Nephrotoxicity in Rats. 3(1), 2011 Jan-Mar, 553-555.

[5]. Sethi P. Momordica charantia L- An Ethnobotanical drug. International Journal of Pharma and Bio Sciences. 3(2), 2012 1-5.

[6]. Waiyaput W, Payungporn S, Issara-Amphorn J, Panjaworayan NT. Inhibitory effects of crude extracts from some edible Thai plants against replication of hepatitis B virus and human liver cancer cells. BMC Complement Altern Med. 12, 2012 246.

[7]. Ganesan A. Natesan S. Perumal PG. Vellayutham R. Manickam K. Ramasamy N. Anxiolytic, Antidepressant and Anti- inflammatory activities of Methanol extract of Momordica charantia Linn. Leaves (Cucurbitaceae). Iranian Journal of Pharmacology and Therapeutics. 7, 2008, 43.

[8]. World Health Organization. The World health report: Mental health: new understanding, new hope. Geneva, 2001. http://www.who.int/whr/2001/en/whr01_en.pdf Accessed on 2012.

[9]. Revised global burden of disease (GBD) 2002 estimates. Geneva, World Health Organization, 2005. http://www.who.int/healthinfo/global_burden_disease/GBD_report_2004update_full.pdf Accessed on 2012.

[10]. O'Donnell JM, Shelton RC. Drug Therapy of Depression and Anxiety Disorders. Brunton LL, Chabner BA, and Knollman BC, Eds., Goodman & Gilman's The Pharmacological Basis of Therapeutics. The McGraw-Hill Companies, New York, USA, 12, 2011, 397-412.
[11]. Nutt DJ. Treatment of Depression. Bennet PN and Brown MJ, Eds., Clinical Pharmacology. Churchill Livingston, Philadelphia, USA, 10, 2008, 331-341.
[12]. Reus VI. Mental Disorders. Longo DL, Kasper DL, Jameson JL, Fauci AS, Hauser SL, Loscalo J, Eds., Harrison's Principle of internal medicine. The McGraw-Hill Companies, New York, USA, 18, 2012, 3536-37.
[13]. Salawu OA, Ezekiel EO, Adaudia O, Kwanashie HO. Toxicological evaluation of Methanolic extract of Momordica charantia leaves in rats. West Afr. J. Pharmacol. Drug Res. 20(1), 2004, 15-21.
[14]. Steru L, Chermat R, Thierry B, Simon P. The tail suspension test: a new method for screening antidepressants in mice Psychopharmacology. 85, 1985, 367–370.
[15]. Porsolt RD, Anton G, Blavet N, Jalfre M. Behavioural despair in rats: a new model sensitive to antidepressive treatments. Eur J Pharmacol. 47, 1978, 379–391.
[16]. Boissier JR, Simon P. Action of caffeine on the spontaneous motility of the mouse. Arch Int Pharmacodyn Ther. 158, 1965, 212–21.
[17]. Porsolt RD, Martin P, Lenègre A, Fromage S, Drieu K. Effects of an extract of Ginkgo biloba (EGB 761) on "learned helplessness" and other models of stress in rodents. Pharmacol Biochem Behav. 36, 1990, 963–971.
[18]. Vogel HG. Drug Discovery and Evaluation Pharmacological Assay Second Edition. Springer-Verlag Berlin Heidelberg, 2002, 570.
[19]. Rang HP, Dale MM, Ritter JM, Flower RJ. Antidepressant drugs.In: Rang HP, Dale MM, Ritter JM, Flower JM. Rang and Dale's Pharmacology. Churchill Livingstone, 6, 2009, 563.
[20]. Krishnan V, Nestler EJ. The Molecular Neurobiology of Depression. Nature. 455, 2008, 894-902.
[21]. Sherman AD, Allers GL, Petty F, Henn FA. A neuropharmacologically- relevant animal model of depression. Neuropharmacol. 18, 1979, 891–893.
[22]. Rang HP, Dale MM, Ritter JM, Flower RJ. Other transmitters and modulators. Rang and Dale's Pharmacology. 6th edition. Churchill Livingstone Publications, 2009, pp 499.
[23]. Pemminati S, Gopalakrishna HN, Shenoy AK, Sahu SS, Mishra S, Meti V, Vinod N. Antidepressant activity of aqueous extract of fruits of Emblica Officinalis in mice. International Journal of Applied Biology and Pharmaceutical Technology. 1(2), 2010, 449-454.
[24]. Chhillar R, Dhingra D. Antidepressant-like activity of gallic acid in mice subjected to unpredictable chronic mild stress. Fundam Clin Pharmacol. 27(4), 2013, 409-18.
[25]. Rocha FF, Lima-Landman MTR, Souccar C, Tanae MM, De Lima TCM, Lapa, Antidepressant-like effect of Cecropiaglazioui Sneth and its constituents – In vivo and in vitro characterization of the underlying mechanism. Phytomedicine. 14, 2007, 396-402.
[26]. Chasapis CT, Loutsidou AC, Spiliopoulou CA, Stefanidou ME. Zinc and human health: an update. Arch Toxicol. 86(4), 2012, 521-34.
[27]. Siwek M, Dudek D, Paul IA, Sowa-Kucma M, Zieba A, Popik P, Pilc A, Nowak G. Zinc supplementation augments efficacy of imipramine in treatment resistant patients: a double blind, placebo-controlled study. J Affect Disord. 118, 2009, 187–195.
[28]. Hall NT, Nagy S, Berry RE. Leaves for Food: Protein and Amino acid contents of leaves from 23 Tropical and Subtropical plants. Florida state Horticultural Society. 1975, 486-490.
[29]. Zhang M, Hettiarachchy NS, Horax R, Chen P, Over KF. Effect of maturity stages and drying methods on the retention of selected nutrients and phytochemicals in bitter melon (Momordica charantia) leaf. J Food Sci. 74(6), 2009, 441-8.
[30]. Aggarwal A, Gaur V, Kumar A. Nitric oxide mechanism in the protective effect of naringin against post-stroke depression (PSD) in mice. Life Sci. 86(25-26), 2010, 928-35.
[31]. Herrera-Ruiz M, Zamilpa A, González-Cortazar M, Reyes-Chilpa R, León E , García MP, Tortoriello J, Huerta-Reyes M. Antidepressant effect and pharmacological evaluation of standardized extract of flavonoids from Byrsonimacrassifolia. Phytomedicine. 18, 2011, 1255– 1261.