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EVALUATION OF ANTIDEPRESSANT, MOTOR COORDINATION AND LOCOMOTOR ACTIVITIES OF HYDROALCOHOLIC EXTRACT OF *BAMBUSA VULGARIS* LEAVES IN MICE

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ABSTRACT

Bambusa vulgaris (*B. Vulgaris*, Poaceae) used in Ayurveda for paralytic complaints, inflammatory disorders and externally to skin disorders. It has various medicinal uses with good nutritional composition and a rich source of vitamins, proteins, amino acid, beta-carotene and phenolic compounds. The aim of present study was to investigate the anti-depressant (by Tail suspension test and Forced swimming test), motor coordination (by Rota-rod method) and locomotor (with an actophotometer) activities of hydroalcoholic leaves extract of *B. Vulgaris* in experimental animals. The study revealed that the extracts (750mg/kg, p.o.) were able to increase the immobility time of rats in a dose-dependent manner when subjected to both Tail suspension and Forced swim tests and the results were similar to that of standard drug Diazepam (2mg/kg, i.p). *B. Vulgaris* hydroalcoholic leaves extract (750mg/kg) showed mild reduction in locomotor and motor coordination activity. Therefore, *B. Vulgaris* may be served as a potential resource for natural psychotherapeutic agent against depression and mood disorders.

Key words: *Bambusa vulgaris*, Tail suspension test, Forced swimming test, Rota-rod method, Actophotometer.

INTRODUCTION:

Depression is a widespread psychiatric ailment (Ferrari et al., 2013). It is already expected to constitute the second largest source of global burden of disease after heart disease in 2020 (Smith et al., 2008). The monoaminergic hypothesis of depression does not provide a full understanding of the progression, causes and pharmacotherapy of depression (Schildkraut et al., 1964). Most accepted hypothesis of depression is postulated and oxidative stress is suggested to be involved in the pathophysiology of depression (Michel et al., 2007). According to WHO estimated, 121 million people suffer from clinical depression (Cryan and Lucki, 2000). It occurs usually in the early adult life of patients with decrease in monoamine neurotransmitters (Dhingra and Sharma, 2005). Medicinal plants therapies may be effective alternatives in the treatment of depression. It possesses least side effects compared to synthetic medicines (Zhang et al., 2015). It has contributed significantly towards the development of modern medicine. Recently, traditional medicine is being re-evaluated by extensive research on different plant species and their active therapeutic principles in worldwide. The rich wealth of plant kingdom can represent a novel source of newer compounds with significant therapeutic activities. The most important merits of herbal medicine seem to be their perceived efficacy, low adverse effects, and low cost (Ghani, 1998). *B. vulgaris* (Poaceae), commonly known as bamboo is taxonomically a grass, but its habit is tree-like. The leaves of *B. vulgaris* revealed that it contains crude protein of 10.1%, phosphorus 86.0mg/100gm, iron 13.4mg/100gm, vitamin B1 0.1mg/100gm, vitamin B_2 2.54 mg/100gm and carotene 12.32mg/100gm (Tamolang et al., 1980). Bamboo leaves have been claimed to be used as an astringent, ophthalmic solution, emmenagogue, vulnerary, and febrifuge to heal the wounds and to control diarrhea in cattle. In ayurvedic medicine, leaves are traditionally used in paralytic complaints and to treat various inflammatory conditions (Kirtikar and Basu, 1990). Evaluation of various plant products according to their traditional uses and medicinal value based on their therapeutic efficacy leads to the discovery of newer and cost-effective drugs for treating various ailments. *B. vulgaris* has an impressive range of medicinal uses with high nutritional value and serves as a good source of vitamins, proteins, amino acid, beta-carotene and various phenolics (Tamolang et al., 1980; Owokotomo et al., 2011). It was reported to possess antimicrobial action (Owokotomo et al., 2011; Senthilkumar et al., 2011), antidiabetic (Senthilkumar et al., 2011), antioxidant (Goyal

et al., 2013), and abortifacient properties (Yakubu and Bukoye, 2009). It has also been reported to possess antidiabetic activity through antioxidant nature. Stem decoction of *B. vulgaris* is used to control menstrual pain (Rai and Nath, 2003). Leaves are used in against fever and diabetes (Pullaiah and Naid, 2003). *B. vulgaris* is used for stomach problems, pain and internal parasites. It is used for skin problems in Trinidad and Tobago (Lans, 2007). In Nigerian folklore medicine, bamboo is used as an emmenagogue, abortifacient, appetizer, and for managing respiratory diseases as well as gonorrhea (Gill, 1992). Leaves and stems are used by the tribes of Raisen, Madhya Pradesh, to treat healing of skin injuries topically. Though the plant and its extracts have been used in the folklore medicine extensively, there is lack of scientific report to support these supposed central nervous system (CNS) depressant activity in mice. The present investigation was undertaken which deals with the evaluation of central nervous system (CNS) depressant activity of hydroalcoholic extract of *B. vulgaris* leaves in mice models.

MATERIALS AND METHODS

Plant material

The leaves of plant *B. vulgaris* were collected from rural area of Bhopal (M.P) in the month of February, 2019. The sample was identified by senior Botanist Dr. Zia-Ul-Hassan, Professor and head of department of Botany, Safia College of Arts and Science, peer gate Bhopal. A herbarium of plants was submitted to the specimen library of Safia College of Arts and Science, peer gate Bhopal and The specimen voucher no. of *B. vulgaris* is 445/Bot./Saf./19. The plant material was dried under shade. It was pulverised to coarse powder with the help of hand grinder. The coarse powder was packed into airtight container and stored in cool and dry place. This material was used for the further study.

Chemical reagents

Imipramine hydrochloride (Sigma-Aldrich, St Louis, USA) and diazepam injection I.P (Ranbaxy Laboratory Ltd., New Delhi, India) were used in this study. All drugs were dissolved in distilled water and administered either intraperitoneal (i.p.) or orally (p.o.). Distilled water was used as the vehicle and all the other chemicals and reagents were of analytical grade and were purchased from S.D. fine Chemicals Pvt. Ltd., Mumbai, India and SRL Pvt. Ltd. (Mumbai, India).

Extraction by maceration method

The shade dried material was coarsely powdered and subjected to extraction with petroleum ether by maceration. The extraction was continued till the defatting of the material had taken place. 50gm of dried plant material

were exhaustively extracted with hydroalcoholic solvent (ethanol: water: 70: 30) using maceration method. The extracts were evaporated above their boiling points and stored in an air tight container free from any contamination until it was used. Finally, the percentage yields were calculated of the dried extracts (Parkhe et al., 2018).

Qualitative phytochemical screening

Crude extracts were screened to identify the occurrence of primary and secondary metabolites, viz. carbohydrates, alkaloids, glycosides, polyphenols, flavonoids, tannins, saponins, terpenoids, proteins and fixed oils, using standard screening test and phytochemical procedures (Pradhan et al., 2019; Harborne, 1973).

Animals

Male and female breed Swiss Albino mice weighing between 25-32 gm were used in the experiments. All the experiments were performed between 9:30 to 16:30 hr to overcome diurnal and circadian variations. All the animals C and in a relative°were housed at a temperature of 24 ± 2 humidity of 65 ± 5%. A 12:12 hr L: D cycle was followed. All the animals were housed in polypropylene cages with paddy husk as bedding with free access to water and fed with standard commercial pelleted chow (Hindustan Lever). All the experimental procedures and protocols used in this study were reviewed by institutional animal ethics committee of Radharaman College of Pharmacy (Proposal number IAEC/RCP/2010/12) and were in accordance with the guidelines of the IAEC.

Acute toxicity studies (LD50)

The acute toxicity study was performed for *B. Vulgaris* (BVHE) using Swiss Albino mice. The animals were fasted for 12 hr prior to the experiment and were administered orally with different dose of BVHE and observed for mortality up to 48 hr (short term toxicity). Based on the short term toxicity, the dose of next animal was determined as per OECD guideline 425, a limit test was performed to categorize the toxicity class of the compound. All the animals dosed at 2000 mg/kg body weight did not show evident toxicity throughout the experimental period. A dose range of 250, 500 and 750 mg/kg was selected for evaluation of pharmacological activities. All the animals were also observed for long term toxicity up to 14 days (OECD, 1996).

Evaluation of antidepressant activity

Tail suspension method

The animals were suspended by a plastic string 75 cm long, about 20 cm above a table top. The duration of immobility was recorded for a period of 6 min (after discarding activity in the first 2 min because animals try to escape during

this period). Mice were considered immobile only when they hung passively and remain motionless. The same procedure was followed animals treated with vehicle, standard drug (diazepam in doses of 2mg/kg, orally) and BVHE 30 min before the test, and percentage change in immobility was calculated (Steru et al., 1985).

Forced swimming test (FST)

The FST is the most widely used pharmacological *in vivo* model for assessing antidepressant activity. Mice were individually placed in cylinder (45×20 cm) containing 15 cm water (25±2°C), so that it could not touch the bottom of the cylinder with its hind limb or tail, or climb over the edge of the chamber. Mice were divided into groups of 5 and received the hydroalcoholic extract aerial parts of BVHE at different doses *viz*. 250, 500 and 750 mg/kg and diazepam (2mg/kg) was used as standard drug. One-hour post administration each mice were placed individually in a tank. Period of immobility (i.e. the total time the animal remained floating in water without struggling and making only those movements necessary to keep its head above water) during the 6 min test period was measured (Porsolt et al., 1977).

Evaluation of motor coordination activity

The motor coordination and performance of each mice was evaluated 1 hr after the extract oral treatment or 30 min after i.p. administration of standard diazepam (2mg/kg) in a Rota-rod apparatus. This equipment has a bar 2.5 cm in diameter and divided into six parts, and it is placed at a height of 50 cm, rotating at 20 rpm. Latency to fall from the rotating bar was registered (Dunham and Miya, 1957).

Evaluation of locomotor activity

The spontaneous locomotor activity of each animal was recorded individually for 10 min using an actophotometer. The movement of the animal cuts off a beam of light falling on the photocell, and the count is recorded digitally. The doses of BVHE (250, 500, and 750mg/kg p.o.) were administered 60 min before the test and diazepam (2mg/kg), used as standard, was given 30 min before the test. The control group was treated with water orally, 60 min before test (Parvathi and Ravishankar, 2013).

Statistical analysis

The values were expressed as mean ± SEM (n=6). The statistical significance was assessed using one-way analysis of variance (ANOVA) followed by Tukey's test and $P<0.05$ were considered to be statistically significant.

Results

The crude extracts so obtained after maceration extraction process was concentrated on water bath by evaporation the solvents completely to obtain the actual yield of extract. The yield of

extracts obtained from the leaves of the plants using hydroalcoholic (Water: Ethanol 30:70) as solvents was found to be 3.8% w/w. The results of preliminary phytochemical screening tests revealed the presence of saponins, steroid, alkaloid and amino acids in the crude extract. The extract BVHE was studied for acute toxicity at doses of 2000 mg/kg b.wt., po. The extract was found devoid of mortality of all animals. So, the doses selected for the antidepressant activity were 250, 500 and 750 mg/kg, po. The BVHE significantly decreased the locomotor activity in a dose dependent manner. The depression of locomotor activity was found maximum at a dose of 750 mg/kg Table 1. BVHE at 750 mg/kg dose caused significant ($p < 0.05$) decrease in fall off time with 78.21% decrement while the standard drug diazepam had fall off time 93.62% decrease (Table 2). Treatment of diazepam showed extremely significant increase (94.71%) in duration of immobility while Imipramine decreased the duration of immobility (71.51%) as tabulated in Table 3. BVHE at dose of 750 mg/kg showed highly significant increase ($p < 0.01$) in duration of immobility (58.97%). Treatment of diazepam showed extremely significant increase (234.38) in duration of immobility while Imipramine decreased the duration of immobility (18.39) as tabulated in Table 4. BVHE at dose of 750 mg/kg showed highly significant increase ($p < 0.01$) in duration of immobility (112.10).

Table 1 Effect of hydroalcoholic leave extract of *B. Vulgaris* on locomotor activity by actophotometer

Group	Dose	Locomotion score (M± SEM)		% Change in locomotion
	(mg/kg, i.p/ p.o)	Basal	After drug administration	
Vehicle control	0.5ml/100gm	492.52 ± 12.19	-	-
Diazepam	2	470.32 ± 12.19	22.26±10.50	95.26
BVHE	250	254.12±11.29	144.09±10.57***	43.29
BVHE	500	315.21 ± 27.31	139.03±30.23***	55.89
BVHE	750	575.11± 45.77	115.65±10.91***	79.89

Values are expressed as mean ± S.E.M with (n = 6) per group. BVTS: *B. Vulgaris* ***$p < 0.001$ compared to standard drug diazepam treated group.

Table 2 Effect of hydroalcoholic leave extract of *B. Vulgaris* on muscle coordination activity by Rota-rod

Group	Dose	Fall off time in sec (M ± SEM)		% Change in fall off time
	(mg/kg, i.p/ p.o)	Basal	After drug administration	
Vehicle control	0.5ml/100gm	199.50 ± 10.00	-	-
Diazepam	2	143.12 ± 13.13	73.92 ± 12.01	93.62
BVHE	250	136.14 ± 13.51	85.36±2.05*	37.29
BVHE	500	146.44 ± 10.20	78.76±4.53^{ns}	46.14
BVHE	750	162.14±12.51	35.32±2.01***	78.21

Values are expressed as mean ± SEM with (n = 6) per group. BVTS: *B. Vulgaris* $^{***}p < 0.001$, $^{**}p < 0.01$ and $^{*}p < 0.05$ and ns = not significant compared to standard drug diazepam treated group.

Table 3 Effect of BVHE on tail suspension induced immobility on mice

Groups	Dose (mg/kg, i.p/ p.o)	Duration of immobility in sec (M± SEM)	% Change in immobility duration
Vehicle control	0.5ml/100gm	125.55 ± 7.23	-
Diazepam	2	246.48±15.30***	94.71
Imipramine	20	13.41±4.95***	71.51
BVHE	250	70.22±7.82^{ns}	30.52
BVHE	500	135.12±10.07 ^{ns}	45.18
BVHE	750	101.12±11.33**	58.97

Values are expressed as mean ± SEM with (n = 6) per group. BVHE: *B. Vulgaris* $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$ and ns = not significant compared to vehicle control group

Table 4 Effect of BVHE on forced swimming induced immobility on mice

Groups	Dose (mg/kg, i.p/ p.o)	Duration of immobility in sec (M± SEM)
Vehicle control	0.5ml/100gm	130.45 ± 6.23
Diazepam	2	234.38±14.30***
Imipramine	20	18.39±3.95***
BVHE	250	65.12±6.82^{ns}
BVHE	500	125.11±9.07^{ns}
BVHE	750	112.10±10.33***

Values are expressed as mean ± SEM with (n = 6) per group. BVHE: *B. Vulgaris* $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$ and ns = not significant compared to vehicle control group

Discussion

Mood disorder is one of the most common mental illnesses with a lifetime risk of 10% in the general population. Most of the drugs that are currently being used in the treatment of depression adversely affect the quality of life of the patient. This leads to patients' noncompliance with medication, which further complicates the problem. Ayurveda, the Indian traditional system of medicine, mentions a number of single and compound drug formulations of plant origin that are used in the treatment of psychiatric disorders (Sembulingam et al., 2013; Pemminati et al., 2012) and are acclaimed to have a lower side-effect profile than conventional drugs. Earlier reports on the chemical constituents of various plants and their pharmacology suggest that plants containing flavanoids and tannins possess activity against many central nervous system disorders (Priyanka et al., 2012). Flavanoids present in the *Hypericum perforatum* (Butterweck et al., 2000) and tannins present in the *Emblica officinalis* (Pemminati et al., 2012) are responsible for antidepressant action. So these components present in the extract may be responsible for the antidepressant action. The FST and TST models of depression are widely used to screen new antidepressant drugs. These tests are quite sensitive and relatively specific to all major classes of antidepressant drugs, including tricyclics, serotonin specific reuptake inhibitors, monoamine oxidase inhibitors and atypical (Dhingra and Sharma, 2005). In the present study, *B. Vulgaris* (250, 500 and 750 mg/kg) produced significant dose-dependent antidepressant effect in behavioural despair tests. Animals subjected to the antidepressant drug treatment struggle more even in desperate situations and they spend less time with immobility (Jayanthi et al., 2012). It has been previously suggested (Reneric and Lucki, 1998) that an increase in both swimming and climbing behaviours in the FST occurs when the animal is treated by a drug which increases serotonin, norepinephrine and dopamine levels in the nerve terminals. An increase in all the three neurotransmitters could be affected by inhibition of monoamine oxidase (MAO) activity in the brain. Tannic acid being a non-selective inhibitor of monoamine oxidase causes an increase in the levels of monoaminergic neurotransmitters in the brain (Pemminati et al., 2012). As the plant *B. Vulgaris* contains tannin and flavonoids, the antidepressant activity may be due to MAO inhibition, thereby increasing norepinephrine and dopamine levels in the brain. Locomotor activity and muscle coordination are an index of alertness and muscle relaxation. Reduction indicates that it may possess a sedative and

skeletal muscle relaxant effect. Decrease in motor activity and muscle relaxation is an indication of CNS depressant property (Leewanich et al., 1996). At a higher dose (750 mg/kg), the extract exhibits a mild sedation and muscle relaxation which may be due to the presence of flavonoids in *B. Vulgaris* and their interaction with the Benzodiazepine site of GABA A receptors (Adeyemi et al., 2006). The mild sedative action of the extract at the higher dose (i.e. 750mg/kg) may be attributed to the presence of some constituents at an optimal concentration to induce sedation. Apart from this, it is of interest to note that several established antidepressants decrease locomotor activity (Hemby et al., 1997). So the sedative effect was not significant when compared with antidepressant activity. Psycho stimulants also reduce immobility in FST and TST models, but in contrast to antidepressants, these cause marked motor stimulation in locomotor activity test. In case of minor or major tranquilizers, immobility was not affected but there was a reduction in motor activity (Dhingra and Sharma, 2005). So it was concluded that antidepressant effect of *B. Vulgaris* was not associated with any motor effects. It confirms the assumption that the antidepressant-like effect of *B. Vulgaris* is specific. Therefore, it was concluded that *B. Vulgaris* may be served as a potential resource for natural psychotherapeutic agent against depression.

Conclusion

Since ancient times, people have been using plants in various ways as a source of medicine. From the above preclinical study, we can conclude that hydroalcoholic leaves extracts of *B. Vulgaris* show a significant antidepressant activity in TST and FST models of depression. We believe that *B. Vulgaris* has the potential to be used as an adjuvant in the treatment of depression and other mood disorders. Further research is required to gain closer insights into the exact mechanism of its action.

References

1. Ferrari, A.J., Charlson, F.J., Norman, R.N., Patten, S.B., Freedman, G., CGL, M., Vos, T., Whiteford, H.A. (2013). Burden of depressive disorders by country, sex, age, and year: findings from the global burden of disease study 2010. PLoS Med. 10(11):1-12.
2. Smith, A.J., Sketris, I., Cooke, C., Gardner, D., Kisely, S., Tett, S.E. (2008). A comparison of antidepressant use in Nova Scotia, Canada and Australia. Pharmacoepidemiol Drug Saf. 17(7):697-706.
3. Schildkraut, J.J., Gordon, E.K., Durell, J. (1965). Catecholamine metabolism in

affective disorders. I. Normetanephrine and VMA excretion in depressed patients treated with imipramine. J Psychiatr Res. 3(4):213-28.

4. Michel, T.M., Frangou, S., Thiemeyer, D., Camara, S., Jecel, J., Nara, K., Brunklaus, A., Zoechling, R., Riederer, P. (2007). Evidence for oxidative stress in the frontal cortex in patients with recurrent depressive disorder-a postmortem study. Psychiatry Res. 151(1-2):145–50.
5. Cryan, J.F., Lucki, I. (2000). Antidepressant like behavioral effects mediated by hydroxy tryptamine receptors. The Journal of Pharmacology experimental Therapeutics. 295:1120-6.
6. Dhingra, D., Sharma, A. (2005). Review on antidepressant plants. Natural Products Radiance.144-52.
7. Zhang. J., Wu, J., Fujita, Y., Yao, W., Ren, Q., Yang, C., Li, S., Shirayama, Y., Hashimoto, K. (2015). Antidepressant effects of TrkB ligands on depression-like behaviour and dendritic changes in mice after inflammation. Int J Neuropsychopharmacol. 1-12.
8. Ghani, A. (1998). Medicinal plants of Bangladesh. The Asiatic Society of Bangladesh: Dhaka.
9. Tamolang, F.N., Lopez, F.R., Semana, J.A., Casin, R.F., Espiloy, Z.B. (1980). Properties and utilization of Philippine bamboos. In: Lessard G, Chouinard A, editors. Bamboo research in Asia. Ottawa: IDRC. 189-200.
10. Kirtikar, K.R., Basu, B.D. (1990). Indian medicinal plants. New Delhi: International Book Distributors. 2724-7.
11. Abidemi, O.O. (2013). Proximate composition and vitamin levels of seven medicinal plants. Int J Eng Sci Invent. 2:47-50.
12. Owokotomo, I.A., Owoeye, G. (2011). Proximate analysis and antimicrobial activities of Bambusa vulgaris L. leaves' beverage. Afr J Agric Res. 6:5030-2.
13. Kennard, W.C., Freyre, R.H. (1957). The edibility of shoots of some bamboos growing in Puerto Rico. Econ Bot. 11:235-43.
14. Senthilkumar, M.K., Sivakumar, P., Faisal, C., Rajesh, V., Perumal, P. (2011). Evaluation of anti-diabetic activity of Bambusa vulgaris leaves in streptozotocin induced diabetic rats. Int J Pharm Sci Drug Res. 3:208-10.
15. Goyal, A.K., Middha, S.K., Sen, A. (2013). Bambusa vulgaris Schrad. ex J. C. Wendl. var. vittata Riviere & C. Riviere leaves attenuate oxidative stress e an in vitro biochemical assay. Indian J Nat Prod Resour. 4:436-40.

16. Yakubu, M.T., Bukoye, B.B. (2009). Abortifacient potentials of the aqueous extract of Bambusa vulgaris leaves in pregnant Dutch rabbits. Contraception. 80: 308-13.
17. Rai, R., Nath, V. (2003). Use of medicinal plants by traditional herbal healers in Central India. In: World forestry congress XII, 21 to 28 September 2003 in Quebec, Canada.
18. Pullaiah, T., Naid, C.K. (2003). Antidiabetic plants in India and herbal based antidiabetic research. New Delhi: Daya Books. 98.
19. Lans, C. (2007). Comparison of plants used for skin and stomach problems in Trinidad and Tobago with Asian ethnomedicine. J Ethnobiol Ethnomed. 3:3.
20. Gill, L.S. (1992). Ethnomedical uses of plants in Nigeria. Benin, Nigeria: Uniben Press. 35-6.
21. Parkhe, G., Jain, P., Jain, D.K. (2018). Total phenolic content, total flavonoid & anti-bacterial activity of hydroalcoholic acacia catechu (l.f.) Willd roots extracts for treatment of mouth ulcer. Pharmacologyonline. 2: 227-233.
22. Pradhan, A., Jain, P., Pal, M., Chauhan, M., Jain, D.K. (2019). Qualitative and quantitative determination of phytochemical contents of hydroalcoholic extract of salmalia malabarica. Pharmacologyonline. 1:21-26.
23. Harborne, J.B. (1973). Phytochemical methods: a guide to modern techniques of plant analysis. 2nd ed. New York: Chapman and Hall.
24. Guideline Document on Acute oral Toxicity Testing, Series on Testing and Assessment No. 423. (1996). Paris: Organization for Economic Co-Operation and Development, OECD Environment, Health and Safety Publications. Available: http//www.oecd.org/ehs.
25. Steru, L., Chermat, R., Thierry B., Simon, P. (1985). The tail suspension test: A new method for screening antidepressants in mice. Psychopharmacology. 85: 367-370.
26. Porsolt, R.D., Bertin A., Jalfre, M. (1977). Behavioral despair in mice: A primary screening test for antidepressants. Arch. Int. Pharmacodyn. Ther. 229: 327-336.
27. Dunham, N.W., Miya, T.S. (1957). A note on a simple apparatus for detecting neurological defi cit in rats and mice. J. American Pharmaceutical Association. 46(3): 208–209.
28. Parvathi M., Ravishankar, K. (2013). Evaluation of Antidepressant, Motor Coordination and Locomotor Activities of Ethanolic Root Extract of Clitoria

Ternatea. Journal of Natural Remedies. 13 (1), 19-24.

29. Sembulingam, K., Sembulingam, P., Namasiyam, A. (2013) Indian J. Physiol Pharmacol. 41: 139-143.
30. Pemminati, S., Gopalakrishna, H.N., Shenoy, A.K., Sahu, S.S., Mishra, S., Meti, V., Vinod, N. (2012). Antidepressant activity of aqueous extract of fruits Emblica off cinalis in mice. Int J. Applied Biology and Pharmaceutical Technology. 1(2):449-454.
31. Priyanka, B., Sridhar, Y., Shankaraiah, P. (2012). Antidepressant and muscle relaxant activity of Cardiospermum halicacabum Linn. Roots in mice. J. Advanced Pharmaceutical Sciences. 2(1):193-200.
32. Butterweck, V., Jurgenliemk, G., Nahrsedt, A., Winterhoff, H. (2000). Flavonoids from Hypericum perforatum show antidepressant activity in the forced swimming test. Planta med. 66(1):3-6.
33. Dhingra, D., Sharma, A. (2005). Evaluation of antidepressant-like activity of glycyrrhizin in mice. Indian J. Pharmacol. 37(6):390-394.
34. Jayanthi, M.K., Prathima, C., Huralikuppi, J.C., Suresha, R.N., Dhar, M. (2012) Anti-deppresant eff ects of Withania somnifera fat (Ashwagandha ghrutha) extract in experimental mice. Int. J. Pharma and Bio Sciences. 3(1): 33-42.
35. Reneric, J.P., Lucki, I. (1998). Antidepressant behavioural eff ects by dual inhibition of monoamine reuptake in the rat forced swimming test. Psychopharmacology. 136:190-197.
36. Leewanich, P., Tohda, M., Matsumoto, K., Subhadhirsakul, S., Takayama, H., Watanbe, H. (1996). Behavioral studies on alkaloids extracted from the leaves of Hunteria zeylanica. Biological and Pharmaceutical Bulletin. 19(3):394-399.
37. Adeyemi, O.O., Yetmitan, O.K., Taiwo A.E. (2006). Neurosedative and muscle-relaxant activities of ethyl acetate extract of Baphia nitida AFZEL. J. Ethnopharmacol. 106(3):316-323.
38. Hemby, S.E., Lucki, I., Gatto, G., Singh, A., Thornley, C., Matasi, J., *et al.* (1997). Potential antidepressant eff ects of novel tropane compounds, selective for serotonin and dopamine transporters. J. Pharmacol Exp Th er. 282: 727-733.