Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

Evaluation of Anti-Depressant Activity of Polyherbal Extract of Hibiscus Rosa Sinesis Flower and Carica Papaya Leaf in Rats

Shweta Singh*, Brijesh Kumar
Shambhunath Institute of Pharmacy, Jhalwa, Prayagraj -211012, (U.P.) India.
Corresponding Author
Shweta Singh
Shambhunath institute of Pharmacy, Jhalwa, Prayagraj-211012 (U.P.) India

(Received: 14 April 2024 Revised: 1 May 2024 Accepted: 18 June 2024)

KEYWORDS

Antidepressant, Forced swim test, Tail suspension test, Hibiscus Rosa sinesis, Carica Papaya, Polyherbal, TLC, Immobility, Herbal drugs, HRSCP.

ABSTRACT

Present study investigated the anti-depressant effects of polyherbal extract of Hibiscus Rosa sinesis flower and Carica papaya leaf in wistar albino rats. The plant material was extracted in a soxhlet apparatus using ethanol as solvent. Preliminary phytochemical screening and thin layer chromatographic analysis was done. Wistar albino rats were selected for experimental study. Imipramine 15mg/kg was used as standard drug for the study. Animals were divided into five groups. For evaluation of anti-depressant effect, tail suspension test and forced swim test method used. HRSCP extract at 400mg/kg was significantly reduced immobility time of animals ($P<0.05$) at day 1st and Day 14th in forced swimming test. Animal treated with HRSCP extract at 200mg/kg significantly reduced immobility time at Day 14th in Tail suspension model ($P<0.01$) as compared to other group. The present study concluded that, polyherbal extract of Hibiscus Rosa sinesis flower and Carica papaya leaf (HRSCP) at dose dependent manner significantly reduced immobility of animals in tail suspension and forced swim test models and produced anti-depressant effects.

According to the findings, Hibiscus Rosa sinensis flower and Carica papaya leaf extract show promise as an adjunct or alternative therapy for depression, maybe providing a low-cost, all-natural option.

Introduction

Plants are the essential needs for our life, either for fulfill oxygen requirements or nutritional requirements. Since past decades plants are used as commercial source of nutrition, medicines and daily requirements[1]. The intricate network of the nervous system enables interaction between an organism and its surroundings. Central nervous system controls functions such as speech, thinking, memory, recognitions movement of skeletal muscle, and involuntary functions such as cardiac function, and controls heart rate, breathing, digestion and endocrine functions etc.[2]. Depression is a common mental condition, characterized by elevated mood and change in behavior, social isolation and loss of interest for long periods involves in activity. About 280 million people (3.8% population) of the world affected from depressive disorder[3, 4].

Hibiscus Rosa sinensis, also known as "Gurhal" is a native plant of India belonging to *Malvaceae* family [5]. *Carica Papaya*, commonly known as Papaya, is a semi wood, tropical fruit plant belonging to *Caricaceae* family[6]. Both of plants having various active phytoconstituents which having different pharmacological activities such as anti-inflammatory, anti-oxidant, anti-depressant etc[7**].** Hibiscus Rosa sinesis flower and Carica Papaya leafs is the plant parts which are utilized since long time ago as alternatives to treat various diseases [8, 9].

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

Materials and Methods

Collection and Authentication of Plant

The plant materials were collected from the local area of Prayagraj, Uttar Pradesh in January 2024. H. Rosa sinesis and Carica papaya, both plants were authenticated at the regional office of Botanical Survey of India, Prayagraj Uttar Pradesh. The plant materials were dried in shade and stored in a paper bag for further used. After submission of both Plant specimen at Botanical Survey of India authentication was done with reference number…..

Chemical and Reagents

All the chemicals and reagents used in the current study were procured from Loba Chemicals Pvt. Ltd. All chemicals employed in the study were analytical grade. The standard drug Hydrocortisone sodium succinate and Imipramine was obtained from Abott Pharmaceuticals as a gift sample.

Animals

Healthy wistar Albino rats of either sex (male or female) weight about (150-180gm) were selected for experimental procedures. The animals were kept in a 12:12 light: dark cycle with a temperature of 22±2 ºC and a relative humidity of 45–55%. The feed pellets and tap water were available to the animals at the time of housing and till experiments.

Physicochemical Evaluation

Plant materials were separately grounded coarsely in a mechanical grinder and after that, materials were subjected for physicochemical parameters such as ash value, total ash, acid insoluble ash, water soluble ash, moisture content and extractive values For determination of purity of crude drug [10].

Preparation of Extract

Both plant materials were grounded coarsely and extracted in a soxhlet apparatus with 95% ethanol used as solvent at 45±5ºC. After extraction the solvent had been evaporated in a rotary evaporator at 40-45ºC. The semisolid mass was obtained after evaporation of solvent at waterbath and stored in a tightly closed container for later use. The extractive value and percentage yield was calculated [10].

Preliminary Phytochemical Investigation

For identification of various active secondary metabolites both of plant extract were studied for various phytochemical tests such as alkaloids, flavonoids, tannins, saponins, sterols etc was performed [11, 12].

Thin Layer Chromatographic Analysis

A TLC plate of 2.5×7.5cm^2 with Silica Gel-G F254 was used for chromatographic analysis of sample. The sample was spotted 1 cm above from the edge of TLC plate. The mobile phase consist of Methanol: Chloroform: water (10: 20:10) is used for running sample along with solvent system. After running solvent to 90% of TLC plate was air dried and observed under UV light at 254nm and 365nm for detection of spots [13].

Grouping of Animals

Total 30 animals were used in the present study. All the animals were acclimatized for seven days before experiments and after that divided into eight groups (n=6). Animals were kept under laboratory condition 22±2ºC with standard food pallet and water ad libitum till completion of experiments with 12 hours light/dark cycle. Animals were divided into five groups; Normal control, Disease control, Standard and two test groups respectively. Each group consists of six animals. All the experimental procedure was performed according to CPCSEA guidelines with approval number-CPCSEA/SIP/006.

Acute Oral Toxicity Study

To assessment of safety profile of extract LD50, acute oral toxicity study was done according to the guidelines demonstrated by OECD 425. Six healthy rats were selected and HRSCP extract at 200mg/kg and 400mg/kg administered orally. Animals were observed for first 24 hours for any toxicity and any psychological symptoms and further for seven days. Observation of mortality of animals was assessed for seven days [14].

Experimental Procedure

Forced Swimming Test (FST)

The forced swimming test was used to describe behavioral activity of animals and method described by Porsolt et al.,1977 with minor modification. Rats were

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

acclimatized for 1 hour before experiments to laboratory condition. Rats were forced to swimming into a glass cylinder (20cm height and 14cm diameter) filled with water with temperature not exceeding 24±1. One day before trial, animals were trained individually for swimming in the glass cylinder to get familiar with experiment and this was called test session. At the day of experiment, animals were received treatment and after 30 minutes placed individually into water container for six minutes for swimming. The swimming time of each rat was recorded. The length of time the mice remained immobile during the final four minutes was noted. On the first, seventh, and fourtenth days of treatment, actual test recordings were made. After six minutes rats were removed from the cylinder and placed into cage[15].

Tail Suspension Test (TST)

Tail suspension test were carried out by methods described by Aslam (2016) with minor modification[16]. After 1 hour of received respective treatments for all groups, animals were subjected for experiments. Rats were acclimatized for 1 hour before experiment to laboratory conditions. This test involved hanging each group of animals from a string that was 58 cm above the table surface and supported by a metal stand. An adhesive tape was placed 1 cm from the tip of each animal's tail. Over the course of a 5-minute observation session, the mice's mobility and immobility were timed for the final four minutes. When mice hang still and passively, they are said to be immobile. Every test animal was kept both visibly and aurally apart from the other animals during the trial. When mice hang still and passively, they are said to be immobile. On days 1, 7, and 14 of treatments, readings were taken [17].

Histopathological Study

At last day of experiments, all animals were sacrificed under light chloroform anesthesia and brain of all animals was dissected out carefully with the help of scalpel and surgical blades. Brain of animals were preserved in 10% formalin buffer and a thin 0.5×0.5cm diameter of brain tissue slide were prepared and stained with hematoxylin and eosin dye solution and observed under light microscope with different magnifications [18].

Statistical Analysis

All the data obtained for different experiments were expressed as Mean± SEM (standard error of mean). Data were analyzed by one way analysis of variance (ANOVA) followed by Tukey's multiple comparison tests for difference among different groups. Data was considered as significant when $P<0.05$.

Results

Physicochemical Analysis

Powdered crude drug material of H. Rosa sinesis and Carica papaya leaf was evaluated for different physicochemical analysis. The result obtained from physicochemical analysis listed below **(Table-8).**

S. No.	Parameter	Result (%w/w)	
		H. Rosa sinesis (flower)	Carica Papaya (Leaf)
1	Total Ash	8.64±0.1	3.22±0.7
2	Water soluble Ash	4.22±0.3	4.67.16±0.1
3	Acid insoluble Ash	2.14±0.2	3.64±0.4
4	Moisture content	6.54±0.1	5.90±0.4
5	Extractive Value	12.60	13.66

Table-1: Physicochemical Analysis of crude drug powder.

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

Preliminary Phytochemical Analysis

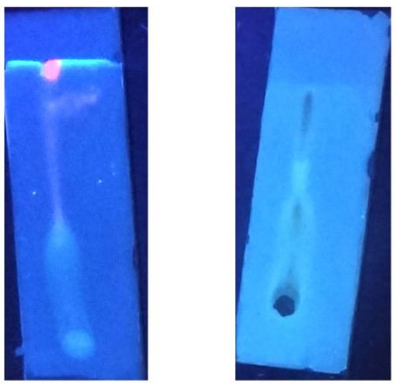
Powdered materials of both plant was extracted individually with 95% ethanol in a soxhlet apparatus after that, reddish brown colored (H. Rosa sinesis) and greenish black (C. papaya) semisolid mass was obtained after evaporation and the % yield was found to 12.6% and 13.66% respectively. The extract of H. Rosa sinesis and Carica papaya had been utilized for determination of different bioactive components of plant. The data obtained from preliminary phytochemical investigation was listed in **(Table-2)**.

S. No.	Phytochemical	Type of Test	Result	
			H. Rosa sinesis	Carica Papaya
1	Alkaloids	Wagner test	+	+
		Mayer's test	-	+
2	Glycosides	Killer-killiani test	+	-
3	Tannins	$Fecl_3$ test	+	+
4	Saponins	Foam test	+	-
5	Flavonoids	Lead acetate test	-	+
		Shinoda test	+	+
6	Phenolics	$Fecl_3$	+	+
7	Sterols	Salkowaski test	-	-
		Lieberman Burchard test	-	-

Table-2: Preliminary Phytochemical Analysis.

Thin Layer Chromatography

Both of plant H. Rosa sinesis and Carica Papaya extract was performed for thin layer chromatography **(Figure-1)** for the identification of bioactive constituents present in plant.



H. Rosa sinesis Carica papaya

Figure-1: TLC Analysis of Plant extract.

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

The Rf values obtained from TLC study mentioned in **(Table-3).** The data obtained from study was compared to previous standard literature and suggested that, both of the plant consist of Alkaloids, Tannins, flavonoids and Phenolic compounds while H. Rosa sinesis having additional glycosides and saponins present. These phytoconstituents possess different pharmacological activity and may have Anti-depressant activity.

S. No.	Result (Rf Values)	
	H. Rosa sinesis	Carica papaya
1	0.22	0.18
2	0.46	0.34
3	0.83	0.51
4	0.94	0.87

Table-3: Rf values obtained from TLC analysis of extract.

Acute Oral Toxicity Study

Acute oral toxicity of HRSCP extract was done in accordance with OECD guidelines and found that, HRSCP extract at 200 and 400mg/kg was found to safe and there is no mortality was seen during the toxicity study.

Anti-Depressant Activity

Forced Swimming Test

Anti-depressant effects of flower of Hibiscus Rosa sinesis and Carica papaya leaf (HRCP) extract against forced swimming test was done and data obtained from animal experiments was listed in **Table-11**. The data obtained from animal study of forced swimming test suggested that, at Day 1, normal saline treated group shows 169.2±11.11, Imipramine 100mg/kg treated animals shows 176 ± 4.54, HRSCP extract 200mg/kg treated animals having 176 ± 6.56 and HRSCP extract at dosage of 400mg/kg having 168.8 ± 8.23 immobility times. It was found that, the immobility time of normal saline treated animals was found greater than HRSCP 400/kg treated animals. Highest immobility time was seen in Imipramine treated animals and HRSCP 400mg/kg treated animals **(Figure-11)**.

Group	Immobility time (sec)	
	Day 1	Day 14
Normal control	169.2±11.11	169.6 ± 7.17
Disease control	196.7 ± 8.32	178.8 ± 7.64
Standard	176 ± 4.54	138.2 ± 7.39
HRSCP extract 200mg/kg	176 ± 6.56	142.6 ± 8.81
HRSCP extract 400mg/kg	168.8 ± 8.23*	109.4 ± 7.80**

Data were expressed as Mean ± SEM, where, $P<0.05$, n=6 for each groups.

Table-4: Data obtained from animal experiment by Forced swimming test in different treatment group of animals. Where, *stand for significant as compared to Imipramine, ** stand for significant as compared to HRSCP 200mg/kg.

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

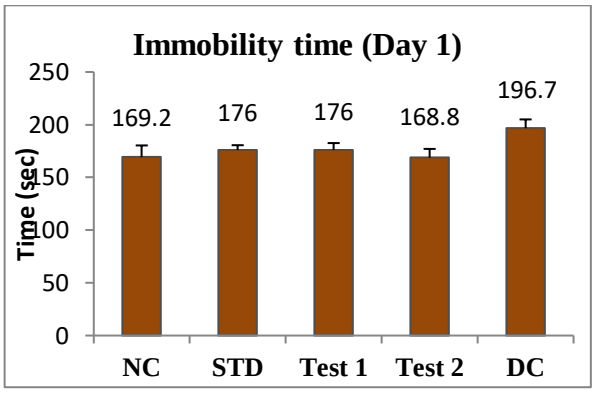


Figure-2: Immobility time of different group of animals by FST at Day-1.

At Day 14, the immobility time recorded through experiment was found that, disease control group animals had been observed for maximum immobility (196.7), normal saline treated animals(NC) having higher immobility time 169.6 ± 7.17 as compared among other group illustrated in **Figure-3**. Imipramine 100mg/kg treated animals was marked for reduced immobility time as compared to normal saline and HRSCP 200mg/kg while less than HRSCP 400mg/kg (109.4 ± 7.80) treated animals.

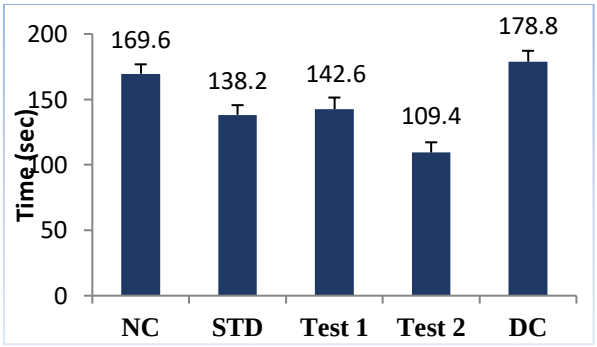


Figure-3: Immobility time of animals in forced swimming test at day -14th

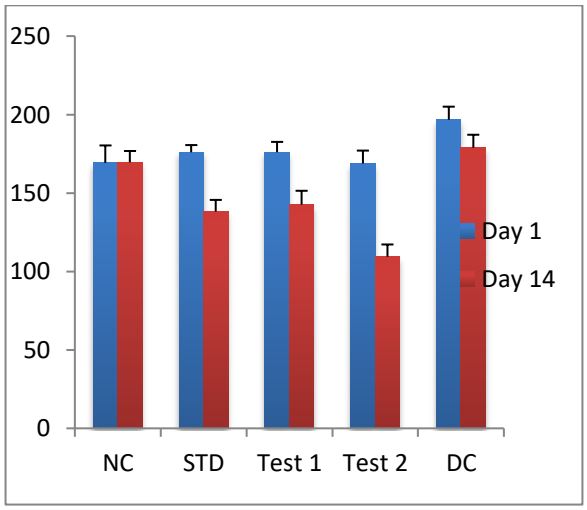


Figure-4: Comparative Anti-depressant effects of different treatments in animal group at day 1st and 14th.

Among all group of animals, HSRCP at 400mg/kg shows significantly reduced immobility time at Day 14th as compared to all groups. Disease control group recorded highest immobility periods at both days. Thus, the figure-13 suggested that HSRCP 400mg/kg extract could reduced immobility time and shows maximum anti-depressant activity in forced swimming test.

Tail Suspension Test

For evaluation of anti-depressant activity, Tail suspension method was also used to monitor animal's immobility time after treatments. **Table-5** showed the immobility time of different group of animals at day-1 and day-14. Maximum immobility was observed in disease control group (236), standard group of animals received Imipramine 100mg/kg had been marked for minimum immobility time (168.0) at day-1.

Treatment Group	Immobility time (sec)	
	Day 1	Day 14
Normal control	207.16 ± 8.97	198 ± 9.47
Disease control	236 ± 9.31	164 ± 14.61
Imipramine 100mg/kg	168 ± 9.78	131.16 ± 8.72

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

HRSCP 200mg/kg	174.66 ± 11.26	116.33 ± 12.34**
HRSCP 400mg/kg	193.5 ± 9.22	133.66 ± 9.17*

Data was expressed as Mean ± SEM, where, n=6, *P*<0.05

Table-5: Immobility time of different group of animal after treatment using Tail suspension methods. Where, * significant as compared to control and ** significant as compared to control, standard and HRSCP 200mg/kg (P<0.05)

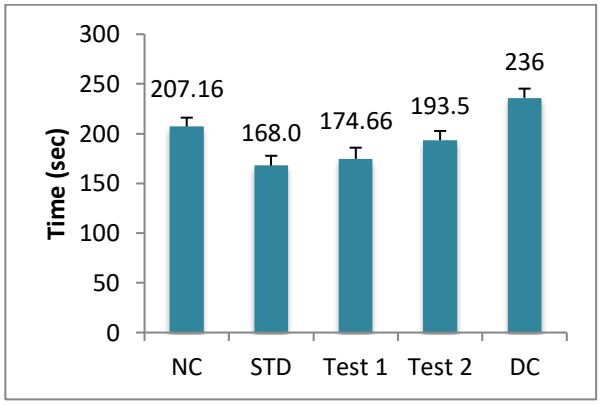


Figure-5: Immobility time of TST of different group of animals at day-1st.

Figure-5 expressed at day 1st after treatment, control group having maximum depression induced as compared to all other groups while standard group showed minimum depression and HSRCP 200mg/kg treated group shows depression less than control (P<0.05) and HSRCP 400mg/kg treated group by tail suspension test

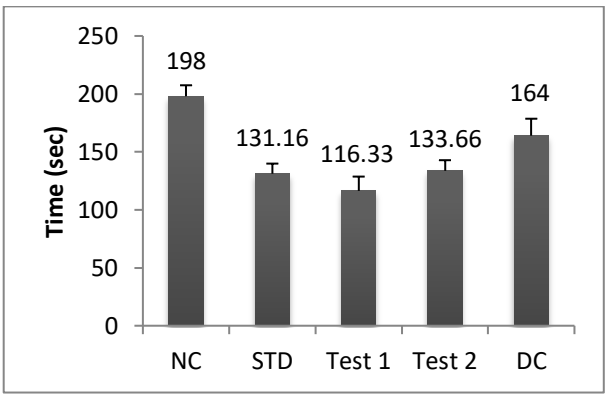


Figure-6: Immobility time of TST after treatment at day-14th.

At Day 14th of TST, the **Figure-6** expressed greater reduction of immobility time in HSRCP 200mg/kg treated animal group. The normal control group shows highest immobility indicated higher depression and Imipramine 100mg/kg treated standard animal group marked for less immobility with reduced depressive symptoms (P<0.05) as compared to normal control and HSRCP 400mg/kg treated animal group.

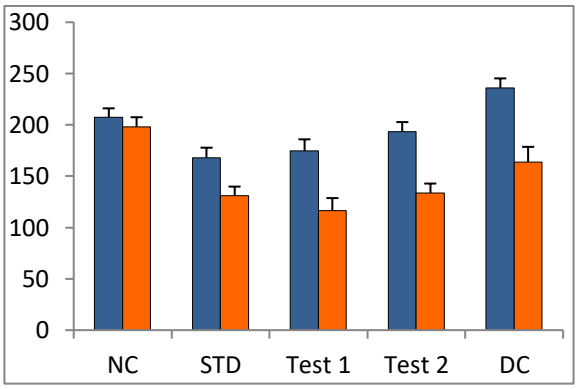


Figure-7: Comparative immobility time of different animal groups by TST at day 1st and Day 14th.

Figure-16 reported that, HRSCP extract at 200mg/kg reduced immobility at 8th day of experiment while, Imipramine 15mg/kg at day 1st observed for reduced immobility as compared to other groups.

Histopahological Study

After completion of experiments, all animals were sacrificed and excised brain for histopathological evaluation of H&E stained brain tissue sections for anatomical changes. **Figure-8** demonstrated the anatomical characteristics of morphological behavior of different animal groups. Normal control (NC) group animal shows normal pattern of cerebral cortex with minute cytoplasm vacuolization and normal anatomical nerve cells. Standard group (STD) animals showed inflamed chromatolysis of depressed animals, mild degenerated neurons.

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

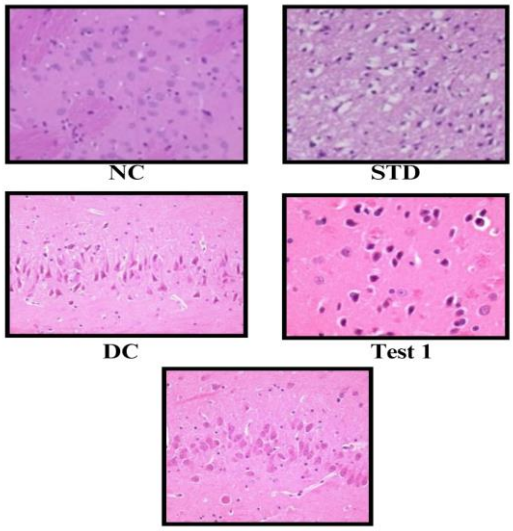


Figure-8: Histopathology of H&E stained brain section of cerebral cortex of experimental animals. Where, NC= normal control, DC= disease control, STD= standard group, Test 1= HRSCP extract 200mg/kg group, Test 2= HRSCP 400mg/kg animal group.

Disease control animals (DC) examined for degenerated nerve cells with acytoplasmic vacuolization in cerebral cortex section. Test 1 group received 200mg/kg polyherbal extract recognized for large acytoplasmic vacuolization and moderately degenerated neurons. While, test 2 (extract 400mg/kg) animals cerebral cortex showed lesser vacuoles with few number of acytoplasmic vacuoles and no degeneration and depressive morphology were observed.

Discussion

Hibiscus Rosa sinesis and Carica papaya are well known Indian plants used traditionally since a long time ago. These plants possess various metabolites such as alkaloids, flavonoids, phenolics and tannins, having a variety of pharmacological activity such as neuroprotective, antidepressant, anti-anxiety etc and were used in treatment of such disorder as herbal remedy [19].

The current study used Wistar albino mice to examine the possible antidepressant effects of a polyherbal extract that combined Hibiscus Rosa sinensis flower and Carica papaya leaf. The current study used Wistar albino mice to examine the possible antidepressant effects of a polyherbal extract that combined Hibiscus Rosa sinensis flower and Carica papaya leaf. The results offer significant perspectives on the medicinal possibilities of natural extracts in the treatment of depression, a common mental health issue.

To assess antidepressant-like effects, the study used well-established behavioural paradigms such the tail suspension test (TST) and the forced swim test (FST). Because of their predictive validity, these tests are commonly used models for screening possible antidepressant drugs in rodents[20].

The active ingredients in the flower and leaf extract of Hibiscus Rosa and Carica papaya may have antidepressant effect because they stimulate the adrenal cortex's glucocorticoid synthesis and release in addition to acting on the GABA/benzodiazepine receptor complex [21].

Anti-depressant effects of HRSCP polyherbal extract was found due to synergistic effects of Hibiscus Rosa sinesis flower and Carica papaya leaf on GABA and Glutamatergic neurotransmission, thus changed behavioral symptoms and reduced immobility time

Conclusion

Data obtained from the study of Anti-depressant activity of polyherbal extract on wistar rats was concluded that, for evaluation of anti-depressant effect, tail suspension test and forced swim test method used. HRSCP extract at 400mg/kg was significantly reduced immobility time of animals ($P<0.05$) at day 1^{st} and Day 14^{th} in forced swimming test. Animal treated with HRSCP extract at 200mg/kg significantly reduced immobility time at Day 14^{th} in Tail suspension model ($P<0.01$) as compared to other group.

The present study concluded that, polyherbal extract of Hibiscus Rosa sinesis flower and Carica papaya leaf (HRSCP) at dose dependent manner significantly reduced immobility of animals in tail suspension and forced swim test models and produced anti-depressant effects. HRSCP polyherbal extract may be used for treatment of depressive disorder and can be a cost effective treatment options today for treatment of depression.

Future research should focus on long-term safety and any drug interactions. The study also evaluated the

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(4), 441-450 | ISSN:2251-6727

safety profile of the polyherbal extract, finding minimal side effects that suggested high tolerance in experimental animals. It takes cautious optimism to translate discoveries from animal models to clinical practice. But according to the findings, Hibiscus Rosa sinensis flower and Carica papaya leaf extract show promise as an adjunct or alternative therapy for depression, maybe providing a low-cost, all-natural option.

References

1. Hashemi SR, Davoodi H. Herbal plants and their derivatives as growth and health promoters in animal nutrition. *Vet Res Commun*. 2011;35(3):169-180. doi:10.1007/s11259-010-9458-2
2. Waugh A, Grant A. Ross & Wilson Anatomy and Physiology in Health and Illness. In: *Ross & Wilson Anatomy and Physiology in Health and Illness*. ; 2018.
3. Goldwaser EL, Aaronson ST. Depressive Disorders. In: *Atlas of Psychiatry*. Springer International Publishing; 2023:531-567. doi:10.1007/978-3-031-15401-0_17
4. Hirschfeld RMA. The Epidemiology of Depression and the Evolution of Treatment. *J Clin Psychiatry*. 2012;73(suppl 1):5-9. doi:10.4088/JCP.11096su1c.01
5. Goldberg KH, Yin AC, Mupparapu A, Retzbach EP, Goldberg GS, Yang CF. Components in aqueous Hibiscus rosa-sinensis flower extract inhibit in vitro melanoma cell growth. *J Tradit Complement Med*. 2017;7(1):45-49. doi:10.1016/j.jtcme.2016.01.005
6. Ali A, Devarajan S, Waly MI, Essa MM, Rahman MS. Nutritional and medicinal values of papaya (Carica papaya L.). *Nat Prod Their Act Compd Dis Prev*. 2012;(August):307-324. doi:10.20959/wjpps20178-9947
7. Sankarganesh P, Joseph B, Kumar G, Illanjiam S, Srinivasan T. Phytomedicinal Chemistry and Pharmacognostic Value of Carica papaya L., Leaf. *J Pure Appl Microbiol*. 2018;12(2):751-756. doi:10.22207/JPAM.12.2.35
8. Zunjar V, Mammen D, Trivedi BM, Daniel M. Pharmacognostic, physicochemical and phytochemical studies on Carica papaya Linn. leaves. *Pharmacogn J*. 2011;3(20):5-8. doi:10.5530/pj.2011.20.2
9. Al-Snafi AE. Chemical constituents, pharmacological effects and therapeutic importance of Hibiscus rosa-sinensis-A review Plants with antioxidant effects View project Plants with antiparasitic effect View project Chemical constituents, pharmacological effects and ther. *IOSR J Pharm www.iosrphr.org*. 2018;8(7):101-119. www.iosrphr.org
10. Seow LJ, Beh HK, Sadikun A, Asmawi MZ. Preliminary phytochemical and physicochemical characterization of Gynura segetum (Lour) Merr (compositae) leaf. *Trop J Pharm Res*. 2013;12(5):777-782. doi:10.4314/tjpr.v12i5.18
11. N Y, M B, J R. Preliminary Phytochemical Analysis of Pentapetes phoenicea L. *IOSR J Pharm Biol Sci*. 2014;9(6):36-39. doi:10.9790/3008-09633639
12. Lakshmi T, Ramasamy R, Thirumalaikumaran R. Preliminary Phytochemical analysis and In vitro Antioxidant, FTIR Spectroscopy, Anti-diabetic activity of Acacia catechu ethanolic seed extract. *Pharmacogn J*. 2015;7(6):356-362. doi:10.5530/pj.2015.6.7
13. Manayi A, Mirnezami T, Saeidnia S, Ajani Y. Pharmacognostical Evaluation, Phytochemical Analysis and Antioxidant Activity of the Roots of Achillea tenuifolia LAM. *Pharmacogn J*. 2012;4(30):14-19. doi:10.5530/pj.2012.30.3
14. OECD. *Test No. 425: Acute Oral Toxicity: Up-and-Down Procedure*. OECD; 2022. doi:10.1787/9789264071049-en
15. Rathod AK, Bapna R, Bundela R, Shukla K. Evaluation of Anti-Depressant Activity on Bryophyllum Pinnatum Leaves. *Int J Pharm Sci Med*. 2023;8(4):1-17. doi:10.47760/ijpsm.2023.v08i04.001
16. Aslam M. Tail suspension test to evaluate the antidepressant activity of experimental drugs. *Bangladesh J Pharmacol*. 2016;11(2):292. doi:10.3329/bjp.v11i2.26517
17. Shashikumara S, C P, Sibgatullah M. Evaluation of Antidepressant Activity of Ethanolic Extract of Alangium Salvifolium Leaves in Swiss Albino Mice. *Biomed Pharmacol J*. 2017;10(1):427-433. doi:10.13005/bpj/1125
18. Pandit A. Ameliorative Effect of Leaves of Carica

papaya in Ethanol and Antitubercular Drug Induced Hepatotoxicity. *Br J Pharm Res*. 2013;3(4):648-661. doi:10.9734/BJPR/2014/4517

19. Kenda M, Kočevar Glavač N, Nagy M, Sollner Dolenc M. Medicinal Plants Used for Anxiety, Depression, or Stress Treatment: An Update. *Molecules*. 2022;27(18):6021. doi:10.3390/molecules27186021
20. Kashmira GJ, Mayuri LA. Rodent Antidepressant Models in Neuropsychopharmacology: A Laboratory Perspective. *Int J Res Pharm Sci*. Published online 2012.
21. Ma Y. Neuropsychological mechanism underlying antidepressant effect: a systematic meta-analysis. *Mol Psychiatry*. 2015;20(3):311-319. doi:10.1038/mp.2014.24