ISSN 2581-6217

World Journal of Pharmaceutical Science & Technology

Journal homepage: www.wjpst.com

Original Research Article

PRELIMINARY PHARMACOLOGICAL SCREENING OF SYNTHESISZED DRUGS FOR ITS ANTIDEPRESSANT ACTIVITY.

Dr. Sunildatta T. Gore[1], Prof. Dr. S.H. Bhosale[2], Mrs. Aditi S. Gore[3], Dr. Abhijeet A Jondhale[4], Prof. Rani J. Gaikwad[5], Dr. Vijay A. Kadnor[6]

1. Professor,at- Usha Dwarkadas Patharikar Institute of Pharmacy, Dogergaon Kawad, Pimpalgaon, Aurangabad.
2. Department of Pharmaceutical Chemistry, Poona College of Pharmacy, Erandwane, Pune-38.
3. Department of Pharmaceutical Chemistry, Poona College of Pharmacy, Erandwane, Pune-38.
4. Assistant Professor, Dr.Kolpe Institute of Pharmacy,Kolpewadi Address-A/P -Nimgaonjali Tahasil- Sangamner Dist-Ahmednagar Maharashtra.
5. Assistant Professor, Arts,Science & Commerce College, Kolhar, Tal-Rahata,Dist.-Ahmednagar.
6. Assistant Professor, Arts,Commerce & Science College, Satral, Tahsil-Rahuri, Dist-Ahmednagar.

Address for correspondence:

Dr. Sunildatta T. Gore, Professor,at- Usha Dwarkadas Patharikar Institute of Pharmacy, Dogergaon Kawad, Pimpalgaon, Aurangabad.
E-mail- sunilgore4u@gmail.com

Received: 15-10-2022, Revised: 29-10-2022, Accepted: 3-11-2022

ABSTRACT

Depressive illness in people is associated with both mental disease and physical signs and changes. The effectiveness plateau experienced with conventional antidepressants, such as monoamine oxidase inhibitors and tricyclic antidepressants, persists despite the extensive use of new antidepressants in clinical settings in recent years. Although animal behavioural models of psychiatric diseases cannot accurately mimic human psychopathology, they can be used to assess the behavioural changes brought on by medications and to propose theories regarding the functioning of the central nervous system and how they relate to mental disorders. This ought to result in a clearer understanding of the therapeutic potential of psychotropic

medications and a more heuristic classification of those substances. Animal models that mimic different elements of depressive diseases are responsive to the antidepressant effects of medications. In order to treat depression more effectively, quickly, with the fewest side effects, and with ease of dosage, a perfect antidepressant must be found. The preliminary pharmacological screening of the synthesised derivatives for the drug's antidepressant action was the following phase, which was then completed.

KEYWORDS: Depression, Antidepression Activity, 2-substituted 1,3,4-oxadiazino[(6,5-b) indole]

INTRODUCTION:

In humans, depressive illness is linked to both mental illness and physical changes and symptoms, including feelings of extreme sadness and hopelessness, mental slowing and memory loss, variable agitation, insomnia, hypersomnia, altered eating patterns, weight loss/overeating, disruption of normal circadian and ultradian rhythms, body temperature, and changes in many endocrine functions.

Animal behavioural models of psychiatric disorders cannot exactly simulate human psychopathology, but they can be used to evaluate the behavioural changes induced by drugs and to suggest hypotheses about the functions of the CNS and its involvement in psychiatric disorders. This should lead to a more heuristic classification of psychotropic drugs and to clarification of their therapeutic possibilities. Various animal models simulate aspects of depressive disorders and are sensitive to the antidepressant effects of drugs.[i] To detect, anticipate, and assess the potential therapeutic effect of psychotropic medicines like antidepressants, numerous animal experimental techniques are employed. Behavioural models, in addition to biochemical and electrophysiological techniques, are crucial in this research. The models are not meant to replicate human psychopathology because human psychiatric diseases cannot be produced in animals, but rather to cause alterations that are responsive to therapeutic drugs in a way that is indicative of how such agents would behave in humans. Behavioural techniques are used to explore antidepressants by inducing behaviour using drugs, lesions, or environments.

Unfortunately, despite the widespread use of novel antidepressants in clinical settings in recent years, the effectiveness plateau seen with traditional antidepressants, such as monoamine oxidase inhibitors and tricyclic antidepressants, remains. New antidepressants should ideally not just start working quickly. The majority of the current antidepressants are anticipated to take between 10 days and 3 weeks to reach clinical efficacy. The search for such an ideal antidepressant as a more effective medication, with better tolerability, rapid onset of action, least side-effects and ease of dosing is required.

Thus, the objective of the research was to develop brand-new tricyclic antidepressants as ß-carboline analogues [pyrido(6,5-b) indoles]. The current compound is intended to potentially block MAO-A

selectively, which is a ß-carboline action. Because MAO-A is inhibited, it is believed that the intended chemical, 2-substituted 1,3,4-oxadiazino[(6,5-b) indole], will have antidepressant effect via raising brain monoamine levels. Accordingly, eight derivatives were synthesized and the preliminary pharmacological screening of these synthesised derivatives for the drug's antidepressant action was the following phase after synthesis of the compound, which was then completed.

Pharmaceutical evaluation for antidepressant activity was done on derivatives of synthetic oxadiazinoindoles, including:

A) The tail suspension test B) A model of the forced swim test C) A quantitative estimation of brain monoamines using the RP-HPLC-FD method.

MATERIAL AND METHOD:

Preliminary Pharmacological Evaluation was carried out using following:

- **Materials**
  - Test compounds 4a-4h were synthesized at Department of Pharmaceutical Chemistry. Poona College of Pharmacy, Erandwane, Pune-38.
  - Noradrenaline bitartrate Salt (Sigma USA)
  - Isoprenaline hydrochloride (Sigma USA)
  - Dopamine HCI injection (Neon Laboratories Ltd. Thane, India)
  - Imipramine HCI (Torrent Pharmaceutical Ltd. Ahmedabad, India)
  - Fluoxetine HCI (Sun Pharmaceutical Ltd. Baroda, India)
  - 5- Hydroxy tryptamine (Sigma USA)
  - Sodium acetate AR (Merck)
  - Ethylene diamine tetra-acetic acid [EDTA] (Qualigens Fine Chemicals)
  - 0-Phosphoric acid (Merck)
  - Perchloric acid (Qualigens Fine Chemicals)
  - Dibutylamine (Qualigens Fine Chemicals)
  - Methanol HPLC (Merck)

- **Animals:**

- Species : Swiss Albino mice were procured from Serum Institute of India, Hadapsar, Pune-24
- Age : Adult
- Weight : 20-25 ± 5 g.
- Gender : Male.
- The animals were housed under 12 hrs day and night conditions.
- The animals have free access to food pellet (Purchased from Amrut laboratory, manufactured by Navmaharasthra Chakan oil mills Ltd, Sangali).
- The Protocol is approved by committee for the purpose of control and supervision of experiments on animals. (CPCSEA).

❖ **Apparatus and instruments:**

- Centrifuge Machine- Eppendorf 5810 R
- Tissue Homogenizer-Remi Equipments Pvt. Ltd.
- Chemical weighing balance-Mettler Toledo, AB204-S Classic, Switzerland.
- Cyclo mixer, CM-101, -Remi Equipments Pvt. Ltd.
- Animal weighing balance-Contech Instruments Co.
- JASCO HPLC system consists of a pump (Model JASCO PU2080, intelligent HPLC pump) and Fluorescence detector (Model JASCO-FP-1520 Intelligent Fluorescence Detector). The Software used was JASCO BORWIN version 1.5, LC-Net II/ADC system.
  - Column Thermo Hypersil C18 ODS (250 mm X 4.6 mm, 5 pm) with Guard Column.

❖ **Preparation of suspension of test drugs:**

- All synthesized compounds practically insoluble in water.
- Suspension of test drugs was prepared by suspending test drug in water containing 2% tween 80 as suspending agent as per S.O.P. no.4

❖ **Preparation of suspension of Imipramine HCl and Fluoxetine:**

Suspension of Imipramine and Fluoxetine was prepared by suspending in water containing tween 80 as suspending agent as per S.O.P. no. **2 & 3.**

❖ **Volume of drug administration:**

The volume of drug solution was calculated based upon the body weight of animal and administered as per OECD guidelines.

❖ **Route of administration:**

The drug solutions were administered orally.

❖ **Pharmacological testing:**

A) Evaluation of antidepressant activity of Test drugs by tail Suspension test:[ii,iii]

Method reported by Steru et al 1985, was followed by S.O.P. no.5

Treatment : 7 days pretreatment

Drugs : Test drugs SHBS1-SHBS8.

Standard drugs : lmipramine HCl and Fluoxetine.

Animals were divided into the following groups

Group 1: Control

Group 2: Test drug 30 mg/kg

Group 3: Test drug 50 mg/Kg

Group 4: Test drug 100 mg/kg

Group 5: lmipramine HCI 15 mg/kg

Group 6: Fluoxetine 20 mg/kg

❖ **Evaluation of Antidepressant activity of test drugs by Despair swim test:[iv][2,3]**

Method reported by Porsolt et al 1977, was followed as per S.O.P. no.6

Treatment : 7 days pretreatment

Drugs : Test drugs SHBSI-SHBS8.

Standard drugs : lmipramine HCI and Fluoxetine.

Animals were divided into the following groups

Group 1: Control

Group 2: Test drug 30 mg/kg

Group 3: Test drug 50 mg/kg

Group 4: Test drug 100 mg/kg

Group 5: Imipramine HCI 15 mg/kg

Group 6: Fluoxetine 20 mg/kg

❖ **Estimation of antidepressant activity of test drug by quantitative estimation of Brain Monoamine levels in mice:**[v]

Method reported by Raju et al (1997) was followed as per S.O.P. no.7

Treatment : 7 days pretreatment

Drugs : Test drugs SHBSI

Standard drugs : Imipramine HC1.

The effect of the test drug SHBS1 (30 mg/kg, 50 mg/kg, 100 mg/kg) on brain monoamine levels of mice brain was studied. Monoamines from brain were isocratically separated and estimated using RPHPLC-FD. The test was used for quantitative comparison of test drugs with the standard drugs.

Animals were divided into the following groups:

Group 1: Control

Group 2: Test drug 30 mg/kg

Group 3: Test drug 50 mg/Kg

Group 4: Test drug 100 mg/kg

Group 5: Imipramine HCI 15 mg/kg

❖ **Acute oral toxicity of Test drugs (1.-D50):**[vi]

LD50 (median lethal oral dose), is a statistically derived single dose of a substance that can be expected to cause death in 50 per cent of animals when administered by the oral route. The LD50 value is expressed in terms of weight of test substance per unit weight of test animal (mg/kg).

Acute oral toxicity of test drugs SHBS1-SHBS8 was carried out according to the OECD guideline by using AOT 425 guideline. Swiss Albino mice (female) weighing between 20-25 g was used.

RESULTS:

- **Acute oral toxicity of SHBSi-SHBS8 Compounds:**

Acute oral toxicity of SHBSI-SHBS8 were carried out according to the OECD guidelines. Results of the acute oral toxicity were shown in table. LD5o of the SHBS1-SHBS8 on statistical estimate based on long term results was found to be greater than 5000 mg/kg for oral administration.

Test type **:** Main test.

Limit dose **:** 5000 mg/kg.

Assumed LD50 **:** none.

Assumed Sigma **:** 0.5 mg/kg

Recommended dose progression **:** 5000, 1750, 550, 175, 55, 17.5, 5.5, 1.75.

- **Acute Oral Toxicity: Short-Term and Long-Term Results:**

Table 1: LD_{50} Data

Code	Sr. No.	Dose mg/kg	Short term result	
			Survived	Died
$SHBS_1$	1	175	6	0
	2	550	6	0
	3	1750	5	1
	4	5500	4	2
$SHBS_2$	1	175	6	0
	2	550	6	0
	3	1750	6	0
	4	5500	5	1
$SHBS_3$	1	175	6	0
	2	550	6	0
	3	1750	6	0
	4	5500	6	0
$SHBS_4$	1	175	6	0
	2	550	6	0

	3	1750	5	1
	4	5500	4	2
$SHBS_5$	1	175	6	0
	2	550	6	0
	3	1750	4	2
	4	5500	4	2
$SHBS_6$	1	175	6	0
	2	550	6	0
	3	1750	5	1
	4	5500	4	2
$SHBS_7$	1	175	6	0
	2	550	6	0
	3	1750	6	0
	4	5500	5	1
$SHBS_8$	1	175	6	0
	2	550	6	0
	3	1750	5	1
	4	5500	4	2

n = 6

Table 2: Tail Suspension Test

Sr.No.	Code	Dose (mg/kg)	Total Immobility time (Sec) Mean ± S.D.	% Decrease in immobility
1	Control	-	142.66(±13.17)	-
2	Imipramine	15	81(±10.71)	43.22
3	Fluoxetine	20	76.33(±12.63)	46.49
4	$SHBS_1$	30	124(±16.19)	13.08
		50	75.66(±15.29)	46.96
		100	126.33(±19.47)	11.44
5	$SHBS_2$	30	132.33(±9.74)	7.23
		50	108.33(±28.71)	24.06

		100	58(±31.03)	59.34
6	$SHBS_3$	30	71.75(±9.51)	49.70
		50	91(_22.31)	36.21
		100	83.75(±27.16)	41.29
7	$SHBS_4$	30	66(±15.13)	53.73
		50	65(±15.43)	54.43
		100	105(±27.31)	26.39
8	$SHBS_5$	30	124.66(±12.43)	12.61
		50	107.66(±14.51)	24.52
		100	78.25 ±18.03	45.14
9	$SHBS_6$	30	123.33(±11.13)	13.54
		50	112.66(±18.61)	21.02
		100	131.5 (±11.41)	7.82
10	$SHBS_7$	30	105.75(±32.43)	25.87
		50	90(±11.40)	36.91
		100	71.25(±21.61)	50.05
11	$SHBS_8$	30	106.66(±10.51)	25.23
		50	98.33(±22.07)	31.07
		100	103(±10.82)	27.80

$n = 6$, $p < 0.05$

Table 3: Despair swim test:

Sr.No.	Code	Dose (mg/kg)	Total Immobility time (Sec) Mean ± S.D.	% Decrease in immobility
1	Control	-	196.5(±5.43)	-
2	Imipramine	15	104.33(±10.06)	47
3	Fluoxetine	20	97.5(±12.65)	48.16
4	$SHBS_1$	30	99.25(±16.45)	49.19
		50	75.00(±21.15)	61.66
		100	105(±14.83)	46.9
5	$SHBS_2$	30	125(±19.74)	36.66
		50	111.5(±21.31)	43.60
		100	58(±31.03)	59.34
6	$SHBS_3$	30	71.75(±9.51)	49.70

		50	91(±22.31)	36.21
		100	83.75(±27.16)	41.29
7	$SHBS_4$	30	66(±15.13)	53.73
		50	65(±15.43)	54.43
		100	105(±227.31)	26.39
8	$SHBS_5$	30	124.66(±12.43)	12.61
		50	107.66(±14.51)	24.52
		100	78.25 (±18.03)	45.14
9	$SHBS_6$	30	123.33(±11.13)	13.54
		50	112.66(±18.61)	21.02
		100	131.5(±11.41)	7.82
10	$SHBS_7$	30	105.75(±32.43)	25.87
		50	90(±11.40)	36.91
		100	71.25(±21.61)	50.05
11	$SHBS_8$	30	106.66(±10.51)	25.23
		50	98.33(±22.07)	31.07
		100	103(±10.82)	27.80

n = 6, $p < 0.05$

BRAIN MONOAMINE LEVEL ESTIMATION:

Table 4: Effect of $SHSB_1$ and Imipramine on levels of Norepinephrine (NE) in mice brain:

Treatment	Sr. No.	Area of NE	Area of IP	Ratio (Y)	NE (ng)/ brain	NE (ng)/ gm tissue
Control	1	309359	19354	15.98424	718.8565	1942.855
	2	396430	17518	22.62987	1033.681	2871.335
	3	304543	18560	16.40857	738.9581	2024.543
	4	389458	16980	22.93628	1048.196	3082.931
	5	360543	18458	19.53316	886.9798	2687.818
	6	345567	16345	21.14206	963.1987	2832.937
					Mean	2574
					Std. Dev	±474.9
Imipramine 15mg/kg	1	435557	84807	5.135861	204.9345	525.473
	2	500741	17151	29.19602	1344.74	3842.115

	3	487599	15564	31.32864	1445.769	4252.262
	4	520805	21907	23.77345	1087.856	3021.822
	5	478562	20902	22.89551	1046.265	2989.329
	6	465781	19583	23.78497	1088.401	3201.181
					Mean	2972
					Std. Dev	±1299
$SHBS_1$ 30 mg/kg	1	433529	17143	25.28898	1159.651	3740.811
	2	452394	19215	23.54379	1076.976	2991.601
	3	408002	19264	21.17951	964.9726	3112.815
	4	421531	16635	25.34001	1162.069	2905.171
	5	423235	16989	24.9123	1141.807	3262.305
	6	409534	17109	23.93676	1095.592	2961.06
					Mean	3162
					Std. Dev	±310.8
$SHBS_1$ 50 mg/kg	1	370729	16517	22.4453	1024.937	3202.929
	2	338615	13647	24.81241	1137.075	3667.983
	3	379375	15367	24.68764	1131.164	2513.698
	4	388918	14209	27.37124	1258.295	3700.867
	5	395143	16780	23.54845	1077.197	3168.226
	6	343789	17854	19.25557	873.8298	2427.305
					Mean	3114
					Std. Dev	±546.6
$SHBS_1$ 100 mg/kg	1	336935	11542	29.19208	1344.554	4337.27
	2	481986	12924	37.29387	1728.361	5083.415
	3	454681	16013	28.39449	1306.769	3843.439
	4	408691	13361	30.58835	1410.699	3358.808
	5	409567	15534	26.36584	1210.666	3026.664
	6	467587	14678	31.85632	1470.767	3001.565
					Mean	3775
					Std. Dev	±820.3

Table 5: Effect of $SHSB_1$ and Imipramine on levels of Dopamine (DA) in mice brain:

Treatment	Sr. No.	Area of DA	Area of IP	Ratio (Y)	DA (ng)/ brain	DA (ng)/ gm tissue
Control	1	332567	19354	17.183373	586.6762	1585.611
	2	316872	17518	18.088366	622.8398	1730.111
	3	320982	18560	17.294289	591.1084	1619.475
	4	318345	16980	18.748233	649.2081	1909.436
	5	312567	18458	16.933958	576.7096	1747.605
	6	299451	16345	18.320649	632.1218	1859.182
					Mean	1742
					Std. Dev	±127.6
Imipramine 15mg/kg	1	204423	84807	2.4104496	-3.65037	-9.35991
	2	291100	17151	16.972771	578.2606	1652.173
	3	210972	15564	13.555127	441.6914	1299.092
	4	285500	21907	13.032364	420.8018	1168.894
	5	215452	20902	10.307722	311.9249	891.2141
	6	205305	19583	10.483838	318.9626	938.1252
					Mean	990.0
					Std. Dev	±561.6
$SHBS_1$ 30 mg/kg	1	217790	17143	12.704311	407.6927	1315.138
	2	229543	19215	11.946032	377.3919	1048.311
	3	175361	19264	9.1030419	263.7859	850.9222
	4	110955	16635	6.6699729	166.5604	416.4009
	5	199456	16989	11.740303	369.1709	1054.774
	6	210435	17109	12.299667	391.5232	1058.171
					Mean	957.3
					Std. Dev	±303.3
$SHBS_1$ 50 mg/kg	1	181551	16517	10.991766	339.2594	1060.186
	2	128844	13647	9.4411959	277.2985	894.5114
	3	169381	15367	11.022386	340.4829	756.6288
	4	239362	14209	16.845802	573.1869	1685.844

	5	210231	16780	12.528665	400.6739	1178.453
	6	198345	17854	11.109275	343.9551	955.4307
					Mean	1089
					Std. Dev	±325.9
$SHBS_1$ 100 mg/kg	1	167637	11542	14.524086	480.411	1549.713
	2	219056	12924	16.949551	577.3327	1698.037
	3	220430	16013	13.76569	450.1055	1323.84
	4	177965	13361	13.319737	432.2852	1029.25
	5	256563	15534	16.516222	560.0169	1400.042
	6	223586	14678	15.232729	508.7284	1038.221
					Mean	1340
					Std. Dev	±269.7

Table 6: Effect of $SHSB_1$ and Imipramine on levels of Serotonin (5-HT) in mice brain:

Treatment	Sr. No.	Area of 5-HT	Area of IP	Ratio (Y)	5-HT (ng)/ brain	5-HT (ng)/ gm tissue
Control	1	268418	19354	13.86886	660.7853	1785.906
	2	187747	17518	10.71738	518.756	1440.989
	3	160456	18560	8.645259	425.3711	1165.400
	4	255129	16980	15.02527	712.9012	2096.768
	5	198567	18458	10.75777	520.5766	1577.505
	6	222498	16345	13.6126	649.2363	1909.518
					Mean	1663
					Std. Dev	±337.1
Imipramine 15mg/kg	1	330225	84807	3.893841	211.2372	541.6337
	2	655442	17151	38.21596	1758.045	5022.986
	3	343249	15564	22.05403	1029.669	3028.439
	4	458976	21907	20.95111	979.9636	2722.121
	5	444510	20902	21.26639	994.1722	2840.492
	6	450342	19583	22.99658	1072.147	3153.375
					Mean	2885

					Std. Dev	±1427
$SHBS_1$ 30 mg/kg	1	453162	17143	26.43423	1227.073	3958.301
	2	342644	19215	17.83211	839.3984	2331.662
	3	399400	19264	20.73297	970.1327	3129.46
	4	328656	16635	19.7569	926.1435	2315.359
	5	345567	16989	20.34063	952.4509	2721.288
	6	361457	17109	21.12672	987.8776	2669.94
					Mean	2854
					Std. Dev	±618.0
$SHBS_1$ 50 mg/kg	1	327554	16517	19.83133	929.4977	2904.68
	2	288545	13647	21.14347	988.6329	3189.138
	3	307679	15367	20.02206	938.0937	2084.653
	4	396639	14209	27.91463	1293.791	3805.268
	5	321872	16780	19.18188	900.2291	2647.733
	6	309561	17854	17.33847	817.1512	2269.864
					Mean	2817
					Std. Dev	±630.4
$SHBS_1$ 100 mg/kg	1	360396	11542	31.22474	1442.969	4654.739
	2	351530	12924	27.19978	1261.575	3710.514
	3	368870	16013	23.03566	1073.909	3158.555
	4	372074	13361	27.84777	1290.778	3073.28
	5	376545	15534	24.24005	1128.188	2820.469
	6	389561	14678	26.54047	1231.861	2514.003
					Mean	3322
					Std. Dev	±764.2

Table 7: Effect of $SHSB_1$ on levels of monoamine in mice brain:

Treatment (mg/kg)	**NE (ng/g of tissue)**	**DA (ng/g of tissue)**	**5-HT (ng/g of tissue)**	**% Increase NE**	**% Increase DA**	**% Increase 5-HT**
Control	1742 ±474.9	1742 ±337.1	1663 ±337.1	-	-	-
$SHBS_1$	3162 ±310.8	957.3 ±303.3	2854 ±618.0	81.51	-40.04	71.61

30 mg/kg						
$SHBS_1$ 50 mg/kg	3114 ±546.6	1089 ±325.9	2817 ±630.4	78.76	-37.48	69.39
$SHBS_1$ 100 mg/kg	3775 ±820.3	1340 ±269.7	3322 ±764.2	116.7	-23.07	99.75
Imipramine 15mg/kg	2972 ±1299	990.0 ±561.6	2885 ±1427	70.60	-43.16	73.48

*negative value indicates % decrease in level as compared to control

DISCUSSION:

The Tail Suspension Test and the Despair Swim Test were two behavioural despair models used to assess the antidepressant efficacy of the produced drugs. It was chosen to conduct a quantitative measurement of brain monoamine levels in mice using RP-HPLC in order to gain insight into the impact of synthetic substances on those levels.

The Tail suspension test was initially used to determine whether the produced drugs had antidepressant properties. The antidepressant effects of all substances were good to moderate and dose-dependent. Imipramine and Fluoxetine, two common medications, reduced immobility by 43.22% (15 mg/kg) and 46.29% (20 mg/kg), respectively. The most effective derivative of the investigated substances, $SHBS_1$, was discovered to cause a 53.73% decrease in immobility at a dose of 30 mg/kg. Other active compounds included $SHBS_3$ and $SHBS_4$., which decreased the immobility time 49.70% and 46.96% and at doses of 30 mg/kg and 50 mg/kg respectively. Other compounds were found to be active only at high dose (100 mg/kg).

Compounds were then put to the Despair swim test. The antidepressant effects of all substances were good to moderate and dose-dependent. Iripramine and fluoxetine, two common medications, reduced immobility by 47.00% (15 mg/kg) and 48.16% (20 mg/kg), respectively. Additionally, $SHBS_1$ was discovered to be the most active in this test, reducing immobility time by 53.73% at a dose of 30 mg/kg. At doses of 50 mg/kg, 30 mg/kg, and 50 mg/kg, respectively, $SHBS_4$, $SHBS_3$, and $SHBS_2$ likewise demonstrated good activity and decreased immobility time by 61.66%, 49.70%, and 43.60%. Other substances only showed notable efficacy at higher doses (100 mg/kg).

According to the findings of behavioural models for antidepressants, $SHBS_1$ showed noteworthy activity. The next interesting step was to use RP-HPLC to measure the compound's impact on the concentrations of monoamines in mice's brains. In comparison to the control, the standard medication imipramine (15 mg/kg) raised the levels of norepinephrine and serotonin by 70.60% and 73.48%, respectively. At a dose of 30

mg/kg, the test medication $SHBS_1$ boosted the levels of norepinephrine by 81.51% and serotonin by 71.81%. Preliminary pharmacological testing has shown that the basic nucleus 1,3,4-oxadiazino[6,5-b]indole does have antidepressant potential, as shown by the capacity of several derivatives to shorten the immobility time in forced swimming and tail suspension tests. Furthermore, since $SHBS_1$ increases norepinephrine and serotonin levels in the brain, it is possible to hypothesise that these substances' MAO-A inhibition is what gives them their antidepressant properties. The analogies between the synthetic compounds and -carbolines, the endogenous MAO-A inhibitor, can be used to support this claim.

CONCLUSION:

According to their capacity to shorten the immobility time in behavioural despair models like the tail suspension test and despair swim test, all derivatives exhibited potential antidepressant action. The most effective derivative in this case, SHBS1, showed considerable antidepressant action in both models even at low dosages. Other noteworthy derivatives are SHBS3 and SHBS4, which in the tested models also shown remarkable antidepressant effect. The lead nucleus, 1,3,4- oxadiazino[6,5-b]indole, was found to have potent antidepressant action in the end. Thus, it would be interesting to carry out detailed pharmacological investigation of these compounds.

REFERENCES:

[i] Thiébot MH, Martin P, Puech AJ. Animal behavioural studies in the evaluation of antidepressant drugs. The British Journal of Psychiatry. 1992 Feb;160(S15):44-50.

[ii] Bch-Rojeckey L.; Kalodera Z.; Samarzija I., The antidepressant activity of Hypericum perforatum L. measured by two experimental methods on mice.Acta Pham. 2004, 54, 157-162.

[iii] Porsolt R D, Rodent Model of Depression: Forced Swimming and Tail Suspension Behavioral Despair Tests in Rats and Mice. Current Protocol in Neuroscience; 2001 , 8.1 OA.1-8.1 OA.IO.

[iv] Porsolt R D., Bwertin A., Jalfre M., Behavioral despair in mice: A primary screening test for antidepressant. Archives International Pharmacodynamics, 1977, 229:327-336.

[v] Madepalli K L., Raju T R., An isocratic assay for norepinephrine, dopamine and 5-hydroxytryptamine using their native fluorescence by high performance liquid chromatography with fluorescence detection in discrete brain areas of rat. Analytical Biochemistry.1997, 246:166-170.

[vi] OECD 425 guideline for testing of chemicals, (Acute Oral Toxicity Up and Down Procedure), 2001.