

# ASSESSMENT OF ANTIOXIDANT, ANTIBACTERIAL, ANTIDEPRESSANT EFFECT AND IN-VIVO BIOCHEMICAL ANALYSIS OF SESBANIA BISPINOSA EXTRACT AND PHYSICAL EXERCISE TREATED STRESS INDUCED EXPERIMENT IN WISTAR RATS

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## ABSTRACT:

**Introduction:** Depression is the most common mental illness and affects more than 10-15% of people. There are a lot of synthetic drugs used to treat depression but these synthetic drugs have potential side effects. Exercise has also been recommended as a complementary therapy that can help improve the symptoms of depressive symptoms and prevent recurrence. **Objective:** The present study aims to assess the antidepressant effect of *Sesbania bispinosa* extracts and Physical exercise in rats and in-vivo Biochemical analysis of the rat serum and also antioxidant, antibacterial activity against leaf extract. **Methods:** The oral administration of extracts, sucrose consumption test was performed to assess the antidepressant activity. After the end of the experimental treatment period [30 days], the animals were sacrificed serum was collected for the biochemical analysis. **Results.** The biochemical parameters were carried out as follows. The serum glucose, triglycerides, Total cholesterol, LDL, was found to be increased in group II depressed rats. The HDL, VLDL, amino acids levels were found to be decreased in group II depressed rats. The case was reversed after the treatment with *S. bispinosa* extract [group IV and V] and in swimming exercise [group VI]. In addition, antioxidant activity results suggested that could be due to polyphenols, but mainly by different molecules or substances present in the extracts and very successful in inhibiting growth of bacterial pathogens. **Conclusion:** *S. bispinosa* extract and Swimming exercise has antidepressant activity, antioxidant, antibacterial activity and this supports its use in ethnomedicine for the treatment of central nervous system disorders.

**Key words:** Antidepressant, *Sesbania bispinosa*, Swimming exercise, lipid profile, Amino acids. Antibacterial activity

## 1. INTRODUCTION

Depression is the world's fourth most serious public health concern, affecting around 350 million people, and is anticipated to become the most frequent mental condition by 2020. [1]. Agitation, nausea, headache, sleepiness or drowsiness, and sexual issues are the most prevalent side effects of these antidepressants [2]. In patients with depression, abnormalities in brain glucose metabolism are prevalent [3]. In situations of depression, the total protein level, as well as the DNA and RNA content, falls dramatically [4]. Few

earlier studies have found that depression is linked to a lipid profile that is abnormal, and that such patients are more violent and suicidal [5]. The Netherlands' Vaan Reed Dortland et al. [6] similarly found an aberrant lipid pattern in depression. Liang Y et al. [7] found high levels of total cholesterol, LDL-cholesterol, and triglycerides in depressed patients.

Amino acids such as tryptophan, phenylalanine, and tyrosine appear to play a role in the aetiology of depressive diseases [8]. The neurotransmitters serotonin and norepinephrine are associated to depression. The amino acids tryptophan and tyrosine are precursors to these neurotransmitters, and MDD sufferers have lower levels of these amino acids [9]. Phenylalanine is used to make tyrosine, the monoamine neurotransmitters dopamine, norepinephrine [noradrenaline], and epinephrine [adrenaline], as well as the skin pigment melanin [10]. All of these substances are essential for the nervous system to function properly.

Since the 1950s, pharmacology has been used to treat depressive illnesses [11]. Herbal therapy is a viable option for treating depression [12]. Herbal remedies are one of the oldest therapies, and plants have long been a source of pharmaceuticals [13]. Over the last decade, there has been a lot of research into finding new therapeutic herbs that can help with depression [14]. Physical activity has been suggested as a supplemental treatment that can help to alleviate depression's residual symptoms and avoid relapse [15]. Exercise therapy's antidepressant impact was published in the British Medical Journal in 2001. [16].

In this study, we investigated the in vivo antidepressant activity of hydroethanolic extract *Sesbania bispinosa* and physical exercise may efficacy to reduce depression. The present study is also undertaken to assess the in vivo potential of *Sesbania bispinosa* and physical exercise by carrying out the various biochemical analysis, antioxidant properties and also assessed on the antibacterial properties of extract *Sesbania bispinosa* against *Escherichia coli*, *Pseudomonas aeruginosa* and *Klebsiella pneumonia* using the agar disk diffusion methods.

## **2. MATERIAL REQUIRED:**

### **2.1. Plant collection and authentication**

The whole plant of *Sesbania bispinosa* were from the local areas [folklore shops] of Coimbatore district, Tamil Nadu, India. The plant was dried in shade at room temperature. The dried whole plant was submitted and authenticated [No.BSI/SRC/5/23/2014-15/Tech-1641] at Botanical Survey of India, Southern Regional Centre, Coimbatore, India.

### **2.2. Procurement of Animals**

Young female Albino rats of Wistar strain [100 ± 20 g] procured from Chettikulam, Nagarcoil, India were used for the study. The Ethical clearance for handling of experimental animals were obtained from the Institutional Animal Ethics Committee

[IAEC] constituted for the purpose and care of laboratory animals as per the guidance of the Committee for the Purpose of Control and Supervision of Experiments on Animals [CPCSEA], Ministry of Social justice and empowerment, Government of India [CPCSEA/No: 264/2015/IAEC].

### **2.3. Grouping of Experimental Animals**

The experimental rats were divided into 6 groups of 6 animals each.

- Group I : Normal control rats
- Group II : CUMS induced depressed rats
- Group III : Experimental rats treated with Imipramine [25 mg/kg]
- Group IV : Experimental rats treated with low dose of leaf extract [250 mg/kg]
- Group V : Experimental rats treated with high dose of leaf extract [500 mg/kg]
- Group VI : Experimental rats treated with swimming exercise

### **2.4. Induction of Depression [17]**

Depression was induced by CUMS protocol in albino rats. Each stress regimen was carried out for two periods with the following stressors; Food deprivation for 24 hours, Day-night reversal, Soiled bedding [ 150ml water per cage] for 22 hours, Cage tilting [ 45 degree inclined] for 22 hours, Crowded housing [10 animals per cage], Exposure to novel odour [household air freshener]

### **2.5. Confirmation of Depression**

#### **2.5.1. Sucrose Preference Test [18]**

The rats underwent adaptive training from day 1 to day 4, with two bottles of pure water provided on days one and two, two bottles of 1% sucrose on day three, and one bottle of pure water and one bottle of 1% sucrose on day four. Rat was given 100 mL of pure water and 100 mL of 1% sucrose solution after 24 hours. The amount of sugar and pure water consumed was recorded.

#### **Calculation**

sucrose preference percentage [%] = sucrose solution consumption [ml]/ [sucrose solution consumption [ml] + water consumption [ml]] × 100%.

#### **2.5.2. Collection of Serum [17]**

After the end of the experimental treatment period [30 days], the animals were sacrificed by cervical dislocation under mild chloroform anaesthesia. Blood was collected by cardiac puncture and the serum was separated by centrifugation at 5000 rpm for 10 minutes.

### **2.6. Chemicals/Reagent kits**

All chemicals and drugs used were obtained commercially and of analytical grade. All the diagnostic kits are products of Sigma-Aldrich Chemicals Pvt Ltd, Karnataka, India.

## 2.7. Biochemical Analysis

Standard sigma commercial kit were used to determine Total glucose, Total protein and lipid profile: Triglycerides, Cholesterol, HDL-Cholesterol, LDL-cholesterol, VLDL-Cholesterol.

### 2.7.1. Determination of Total Glucose Concentration

Glucose estimation was done by the glucose oxidase method and light absorbance was absorbed via colorimeter at 546nm.

### 2.7.2. Determination of Total Protein Concentration

Total protein concentration is measured using the Biuret reagent. Kit comprises of copper II [cupric ion] alkaline solution, which interacts with the peptide bond to form a blue adduct which is measured at 540nm.

### 2.7.3. Determination of Total Cholesterol

Cholesterol esterase hydrolyzes cholesterol esters in the blood [CHE]. Cholesterol oxidase [CO] converts free cholesterol to cholest-4-en-3-one, which produces hydrogen peroxide [H<sub>2</sub>O<sub>2</sub>, which oxidatively interacts with 4-aminoantipyrine and phenol in the presence of peroxidase [POD] to produce a red chromophore. The red quinoneimine dye produced is spectrophotometrically measured at 505 nm.

### 2.7.4. Determination of Triglycerides

Triglycerides are determined after enzymatic hydrolysis with lipase. Lipases hydrolyze serum triglycerides into glycerol and free fatty acids. Glycerol is transformed to glycerol-3-phosphate [G -3-P] in the presence of ATP and glycerol kinase, which is then oxidised by GPO to produce hydrogen peroxidase. Peroxide catalyses the formation of a coloured quinoneimine complex detectable at 546 nm from hydrogen peroxide, 4-amino antipyrine, and ESPAS.

### 2.7.5. Determination of HDL – Cholesterol

HDL – Cholesterol reagent reacts directly with LDL and VLDL at pH 10 to form insoluble complexes. This action occurs at room temperature. Centrifugation was used to remove the precipitate, and the supernatant is tested for HDL cholesterol.

### 2.7.6. Determination of LDL – Cholesterol

Friedwald's formula can be used to calculate LDL cholesterol.

$$LDL = TC - \frac{[HDL + Triglyceride]}{5}$$

### 2.7.7. Determination of VLDL - Cholesterol

The following formula can be used to determine VLDL - Cholesterol.

$$\text{VLDL} = \frac{\text{Triglyceride}}{5}$$

### 2.7.8. Determination of aminoacids

The concentration of amino acids such as phenylalanine, tyrosine, and tryptophan was determined using the method reported by Bhaska, 2014 [19].

### 2.8. Determination of phenylalanine

To the testtube, add 0.2ml of serum sample. In that, added 2 ml of nitration mixture [10g KNO<sub>3</sub>/ 100ml H<sub>2</sub>SO<sub>4</sub>], kept for waterbath for 30 minutes and add 5ml hydroxylamine ammonium sulphate mixture[15g NH<sub>2</sub>OH.HCl + 20g [NH<sub>4</sub>]<sub>2</sub>SO<sub>4</sub> / 100ml H<sub>2</sub>O], After 5 minutes of incubation at 37°C, add 2 mL of a 20% NaOH solution. On an ice bath, the contents were incubated for 10 minutes. In a colorimeter, the colour developed was measured at 520 nm. Distilled water was taken as blank, Standards in the range of 200 – 1000 µg were taken and treated in a similar manner.

### 2.9. Determination of Tyrosine

To the testtube, add 0.2ml of serum sample. In that, added 1 ml of nitrosonaphthol reagent [15% w/v in 0.1 N NaOH] and add 2 ml Acid-Base mixture [equal volume 0.025 N HNO<sub>3</sub> + 0.3 N NaOH ], kept for incubation for 10 minutes in waterbath, then add 4ml H<sub>2</sub>SO<sub>4</sub> dropwise. Measure the red colour developed was read at 520 nm against blank in colorimeter. Distilled water was taken as blank, Standards in the range of 30 – 150 µg were taken and treated in a similar manner.

### 2.10. Determination of Tryptophan

To the testtube, add 0.2ml of serum sample. In that, added 1 ml of Ehrlich reagent [3% w/v in 2 N HCl] and add 8 ml H<sub>2</sub>SO<sub>4</sub> [23.7 N], kept for incubation for 1 hour at room temperature. then add 0.1 ml NaNO<sub>3</sub>. The contents were incubated for 10 minutes at 37°C. Measure the colour developed was read at 520 nm against blank in colorimeter. Standards Tryptophan in the range of 20 – 100 µg were taken and treated in a similar manner.

### 2.11. Scavenging Activity of DPPH Radical

DPPH radical scavenging activity was determined according to [21] with some modifications. The reaction mixture consisted of 0.5 mL of *S. bispinosa* plant extract, 3 mL of methanol, and 0.5 mL of 0.5 mM 2,2-diphenyl-1-picrylhydrazyl [DPPH] radical solution in methanol. After incubation for 45 min, absorbance was determined in a spectrophotometer at 517 nm.

The antioxidant activity was calculated by using the following equation.

$$\% \text{ inhibition} = \frac{[A \text{ control} - A \text{ sample}]}{A \text{ control}} \times 100$$

A control = Absorbance of negative control at the moment of solution preparation  
A sample = Absorbance of sample after 45 min

### 2.12. Antibacterial activity of *Sesbania bispinosa* extract

Experimental bacteria were purchased from Microbial facilities at GBIMT, Chandigarh, India. The antibacterial activity of *Sesbania bispinosa* extract against harmful pathogens such as *P. aeruginosa*, *E. coli* and *K. pneumonia* was tested by the standard Kirby–Bauer disk diffusion method [22]. To achieve this method, Mueller-Hinton agar was autoclaved solution transfer to the Petri plates. Those plates were sterilized under UV light. Bacterial cultures were swabbed into MHA plates and sterilized filter paper disks 6 mm were placed on MHA plates. Then the disks were loaded with distilled water as a control, *Sesbania bispinosa* extract [different sample dose 50µg/ml, 100µg/ml and 150µg/ml] were separately placed on the media. The bacterial inoculated plates were incubated at 37°C for 24 h. The diameters of the zones of bacterial growth inhibition surrounding were measured using Hi-Media antibiotic zone scale.

### 2.13. Statistical Analysis

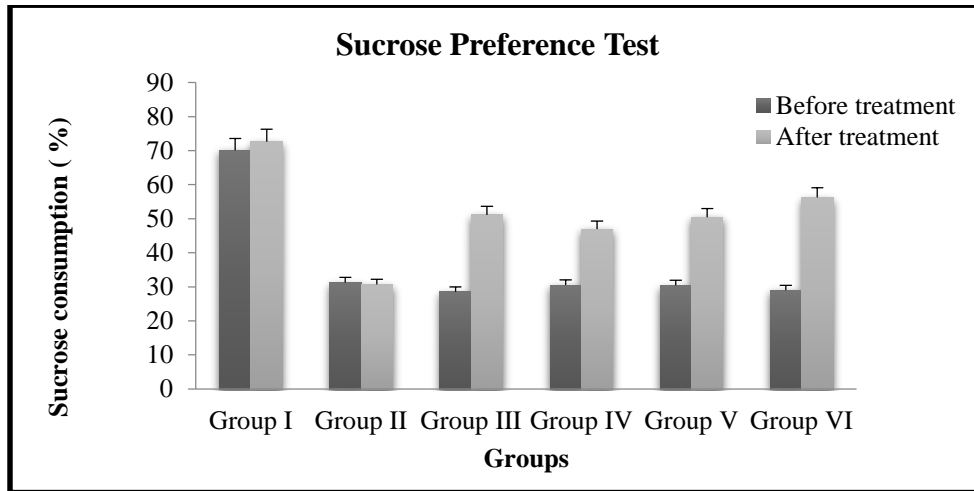
All values were expressed as Mean ± S.E.M. The findings were statistically evaluated by one-way ANOVA, finding  $P < 0.05$  to be significant.

## 3. RESULT AND DISCUSSION

### 3.1. Sucrose Preference Test:

The SPT [Sucrose Preference Test] was a reward-based test that was used to confirm depression. Sweet meals or solutions pique the curiosity of rodents from birth. Reduced preference for sweet solution in SPT indicates depression, which can be reversed with antidepressant medication [23].

As shown in Figure 1, when wister albino rats were exposed to the CUMS technique, the percentage of sucrose consumed in the stressed rats was much lower than in the control animals. However, post-hoc analysis revealed that long-term treatment of stress induced albino rats with plant extract and swimming exercise groups were improved sucrose consumption, as compared to depressed rats [group II]. After treatment, the study found that a high dose of *S. bispinosa* produces higher antidepressant activity than a lower dose of plant extract.



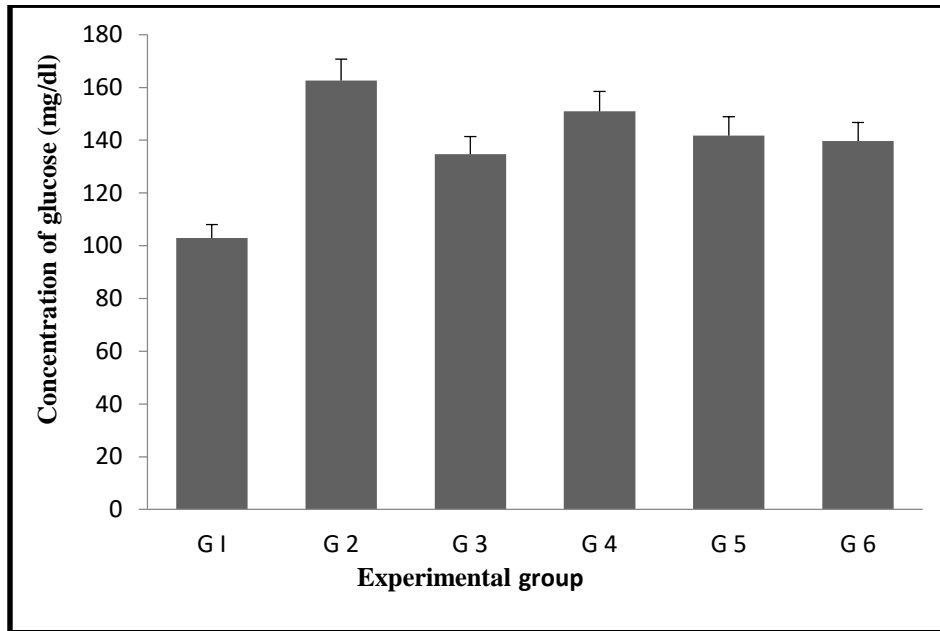
**Figure 1: Sucrose Preference Test**

The induction of major depression in humans was modeled by causing a reduction in reward responsiveness, as evidenced by decreased intake of sucrose solutions [23]. The results of this study are reliable with previous research that indicate a substantial decrease in the proportion of sucrose consumed by rats [24]. Swimming exercise substantially overturned this behavioral shift as compared to *S. bispinosa* extract, implying that it has an antidepressant-like result.

### 3.2. Biochemical Analysis

#### 3.2.1. Determination of Total Glucose concentration:

Figure 2 shows the glucose levels of normal and experimental rats after 30 days of treatment. In normal control group [group I], serum glucose levels were near 100 mg/dl. Glucose levels in the depression-induced group [group II] were higher than 100 mg/dl. In comparison to normal control rats, oral treatment of imipramine [group III] generates a considerable response in Wistar rats, with a significant drop in glucose level. The depression control was dramatically reduced after treatment with a hydroethanolic extract of *S. bispinosa* [group IV & V]. The exercised groups [group VI] were compared to their equivalent groups, and the results were similar to those of standard medications [group III].



**Figure 2: Total Glucose Concentration In The Serum**

Glucose is the primary energy source for brain cells, and the glucose transporter [GLUT] family is responsible for its entry [25]. In the human brain, glucose transporters 1 [GLUT1] and 3 [GLUT3] are primarily transmembrane glucose transporters. [26].

DNA methylation of the main promoter regions of GLUT1 was much higher in depression patients' brain cells than in healthy comparison subjects, lowering the efficacy of GLUT1 to absorb glucose from blood vessels to cells and compromising brain metabolism. After depression patients were treated, DNA methylation of the GLUT1 promoter was significantly reduced. This could indicate that GLUT1 increases are linked to good depression treatment [27]. Li et al., 2020 and Linda et al., 2011[28&29] both published similar reports. Their research findings show that antidepressant effects are linked to glucose metabolism

### **3.2.2. Determination of Total protein triglycerides and cholesterol in the serum:**

The present study shows the estimation of serum protein of rats. In depressed rats group II, the levels of total protein was significantly decreased when compared to that of normal control rats [group I]. After treatment, the total protein levels were found to be reversed. The standard drug Imipramine treated rats [group III] showed increased level of protein than the depressed rats. The low and high dose plant extracts [group IV & group V] revealed the increased levels of protein than the depressed rats. The high dose of hydroethanolic extract of *S. bispinosa* shows higher activity than the exercised group [group VI].



Protein and hence specific amino acids, can have an impact on brain function and mental wellness. Amino acids are used to make many neurotransmitters in the brain. The amino acid tyrosine is used to make the neurotransmitter dopamine, while tryptophan is used to make the neurotransmitter serotonin [30]. There will be insufficient synthesis of the respective neurotransmitters if any of these two amino acids is deficient, which is linked to low mood and violence in people. Thus the study shows significant difference between each group that is statistically determined as shown in **Table 1**.

**Table 1. Concentration of total protein triglycerides and cholesterol in the serum**

Groups	Protein [ $\mu\text{g}$ ]	Triglycerides [mg/dl]	Cholesterol [mg/dl]
Group I	101.66 $\pm$ 1.6 <sup>a</sup>	77.61 $\pm$ 0.57 <sup>b</sup>	151.23 $\pm$ 1.94 <sup>e</sup>
Group II	53.08 $\pm$ 1.99 <sup>f</sup>	85.88 $\pm$ 1.87 <sup>a</sup>	182.99 $\pm$ 1.91 <sup>a</sup>
Group III	91.43 $\pm$ 1.44 <sup>b</sup>	75.41 $\pm$ 1.86 <sup>bc</sup>	160.25 $\pm$ 2.25 <sup>d</sup>
Group IV	66.07 $\pm$ 1.77 <sup>e</sup>	78.19 $\pm$ 1.45 <sup>b</sup>	176.52 $\pm$ 1.31 <sup>b</sup>
Group V	72.94 $\pm$ 1.65 <sup>d</sup>	74.86 $\pm$ 1.72 <sup>c</sup>	166.17 $\pm$ 1.88 <sup>c</sup>
Group VI	83.12 $\pm$ 1.67 <sup>c</sup>	76.03 $\pm$ 1.22 <sup>bc</sup>	163.07 $\pm$ 2.12 <sup>cd</sup>
F	270.42	247.36	212.56
P-value	6.73	6.57	5.78

Data represent mean [n = 3]  $\pm$  SE within each column means with different superscript letters are statistically significant at P<0.05.

### 3.2.3. Determination of Lipid profile:

The current work examines the estimation of rat serum lipid profiles under various conditions. When compared to normal control rats, the levels of Triglycerides, Total cholesterol, and LDL were considerably higher in depressed rats [group II]. The levels of HDL and VLDL in group II were substantially lower than in group I normal control rats. Triglyceride, total cholesterol, HDL, LDL, and VLDL levels were observed to be reversed after treatment. Triglycerides, total cholesterol, and LDL levels were lower in rats treated with the conventional medication Imipramine, but HDL and VLDL levels were higher, similar to normal control rats. Treatment with a high dose of plant extract has the same

effect as regular medications. The exercise treated rats [group VI] reveal a statistically significant difference with normal and depression control rat. Thus the study shows significant difference between each group that is statistically determined as shown in **Table 2** Similar results were reported by Umadev et al., 2010 using *C. pepo* seed extracts in rats [31].

Individuals with depression had slightly higher total and LDL cholesterol readings [32] These findings are consistent with two recent studies that demonstrated greater TG, total and LDL cholesterol levels, as well as lower HDL cholesterol levels, in people with depression compared to healthy controls [33]. In depressed people, one study found that their TG and HDL cholesterol levels were both greater [34] Individuals with serious depression had considerably higher TG levels, which were also positively linked with disease severity in this study [35].

**Table 2. Lipid profile in the serum**

Groups	HDL [mg/dl]	LDL[mg/dl]	VLDL[mg/dl]
Group I	85.42±1.34 <sup>a</sup>	51.09±1.96 <sup>e</sup>	15.02±1.70 <sup>a</sup>
Group II	43.04±1.92 <sup>e</sup>	75.47±1.59 <sup>a</sup>	9.77±1.90 <sup>d</sup>
Group III	72.58±1.34 <sup>b</sup>	60.04±1.97 <sup>d</sup>	14.42±1.57 <sup>a</sup>
Group IV	54.62±1.39 <sup>d</sup>	70.86±2.06 <sup>b</sup>	11.08±1.83 <sup>cd</sup>
Group V	63.69±1.71 <sup>c</sup>	65.46±1.26 <sup>c</sup>	12.85±2.08 <sup>c</sup>
Group VI	61.28±1.90 <sup>c</sup>	67.39±1.34 <sup>bc</sup>	14.01±1.85 <sup>b</sup>
F	485.62	147.54	78.51
P-value	9.61	3.52	4.57

Data represent mean [n = 3] ± SE within each column means with different superscript letters are statistically significant at P<0.05.

Depression has been linked to increased sympathetic nervous system activity and the hypothalamic-pituitary-adrenal cortical axis [36]. Cortisol raises serum levels of circulating free fatty acids, which stimulates the synthesis of very-low density lipoprotein [VLDL] in the liver, resulting in higher TG levels [37]. Some studies have found that depressed persons have greater amounts of very low density lipoprotein [VLDL] in their blood [38], while others have found that depressed people have higher overall cholesterol levels [39]. Depressed people exhibited higher amounts of LDL and its apolipoprotein B [apoB, a constituent of LDL] in their blood than healthy people, but lower levels of HDL and its apolipoprotein A [apoA, a part of HDL] [40].

### 3.2.4. Determination of Aminoacids:

Depression-induced wister albino rats were treated with imipramine, a hydroethanolic extract of *S. bispinosa*, and swimming exercise. Table 3 shows the effect of different dosages of *S. bispinosa* extracts on serum amino acids such as phenylalanine, tyrosine and tryptophan levels. The level of amino acids in depressed rats [group II] was lower. The hydro ethanolic extract of this plant's leaves increased amino acids levels, indicating that it possesses antidepressant properties. Following the exercise treatment, group VI increased their levels of amino acids, which has depressive properties. Because exercise had a similar effect on the imipramine-treated group [group III]. With  $p < 0.05$ , both the exercised and hydroethanolic *S. bispinosa* extract treatment groups differ statistically from the normal and depressive control rats.

**Table 3. Aminoacids profile in the serum**

Groups	Phenylalanine [mg/dl]	Tyrosine [ $\mu$ g/dl]	Tryptophan [mg/dl]
Group I	1.46 $\pm$ 0.57 <sup>a</sup>	26.32 $\pm$ 1.54 <sup>a</sup>	22.52 $\pm$ 1.09 <sup>a</sup>
Group II	0.67 $\pm$ 0.09 <sup>d</sup>	13.94 $\pm$ 1.66 <sup>e</sup>	10.39 $\pm$ 1.18 <sup>e</sup>
Group III	1.33 $\pm$ 0.11 <sup>ab</sup>	21.05 $\pm$ 1.70 <sup>ab</sup>	17.91 $\pm$ 1.55 <sup>b</sup>
Group IV	0.80 $\pm$ 0.07 <sup>c</sup>	15.68 $\pm$ 1.91 <sup>d</sup>	12.78 $\pm$ 1.67 <sup>d</sup>
Group V	0.96 $\pm$ 0.06 <sup>bc</sup>	17.62 $\pm$ 1.75 <sup>c</sup>	13.01 $\pm$ 0.84 <sup>cd</sup>
Group VI	1.05 $\pm$ 0.04 <sup>b</sup>	19.04 $\pm$ 0.78 <sup>b</sup>	15.98 $\pm$ 1.69 <sup>c</sup>
F	9.17	57.76	59.66
P-value	2.21	1.72	1.11

Data represent mean [n = 3]  $\pm$  SE within each column means with different superscript letters are statistically significant at  $P < 0.05$ .

Lower amounts of phenylalanine amino acids were found to be key contributors in the pathophysiology of depression, confirming prior findings [41]. The serum content of phenylalanine was considerably lower in MDD patients, according to Islam et al [42]. Catecholamine precursors [phenylalanine, tyrosine] have been recommended as a viable treatment for depression in various researches [43]. Phenylalanine has a number of beneficial effects as a nootropic, including better motivation, increased concentration and focus, anxiety alleviation, and mood enhancement [44].

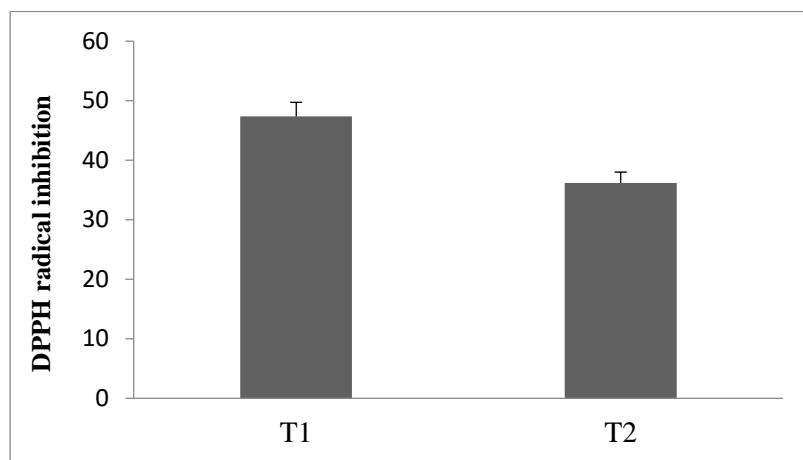
Tyrosine was shown to be a precursor to the neurotransmitters adrenaline, dopamine, and norepinephrine, and it has been linked to depression [46]. However, tyrosine has been proven to be beneficial in a number of studies, including stress in mice, long-term labor and sleep deprivation [47], stress hormone decrease and benefits in cognitive and

physical performance [48]. There have been a few instances suggesting tyrosine metabolism in depressed people was aberrant [49]. Kishimoto and Hama found that depressed patients' tyrosine levels were much lower than controls', and that tyrosine levels rose as the patients recovered from their depression [50].

Tryptophan was the only precursor to the neurotransmitter serotonin, which was synthesised both peripherally and centrally [51]. Because tryptophan was transformed to 5-hydroxytryptophan [5-HTP], which was subsequently converted to the neurotransmitter serotonin, it has been suggested that taking tryptophan or 5-HTP can help with depression symptoms by raising serotonin levels in the brain [52]. The tryptophan depletion model has been utilised in clinical and preclinical studies to test the concept that reduced serotonin production is linked to depression [53]. According to another study, there was no link between tryptophan intake and depression [54]. Tryptophan was a substrate for the vast neutral amino-acid transporter system, and it competes with numerous other important amino acids for brain function for transport [55].

### 3.3. Scavenging Activity of DPPH Radical

Results were detected that the DPPH radical scavenging activity was positively correlated to the concentration of the extract.  $EC_{50}$  values of the extracts evaluated in this study are shown in [Fig 3].



Values are the average of three replicates  $\pm$  SE and expressed in  $EC_{50}$  temperature basis. Means followed by different letters are significantly different [ $p \leq 0.05$ ]. T1 = extract obtained at room temperature, T2 = extract obtained by boiling

#### Figure 3. DPPH radical scavenging activity of *S. bispinosa*

In general, extracts of room temperature methanol extract samples showed higher DPPH radical scavenging activity, when compare to boiling extract samples, which they showed the lowest values of  $EC_{50}$  [47.35  $\pm$  1.03 and 36.18  $\pm$  1.48 mg GAE/L], respectively. DPPH radical scavenging activity, of methanol extract of *A. bisporus*, *P. dryinus*, *Boletus edulis*,

and *P. ostreatus* with  $EC_{50} = 78.43, 58.06, 38.31,$  and  $29.66$  mg GAE/mL, respectively [56]

### 3.4. Antibacterial properties

The antibacterial activities of the *S. bispinosa* extract were performed against *P. aeruginosa*, *E. coli* and *K. pneumonia* by disk diffusion assays. The diameter of the inhibition zone of Control 50µg/ml, 100µg/ml and 150µg/ml is shown in Table 4. This result indicates that no antibacterial activity was observed with Control against three bacterial strains and high concentration of 150 µg/ml exhibited the highly inhibitory action opposition to *P. aeruginosa*, *E. coli* and, *K. pneumonia* was 14.2 mm, 13.4 mm, and 16.4 mm respectively, when compared to low concentration

**Table 4: Inhibition zone induced by extract of *Sesbania bispinosa* leaves against bacterial pathogens**

Values are mean ± standard deviation of three replicates

The mean diameter of inhibitory zone [mm]				
Bacteria	Control	Sesbania bispinosa extract		
		[50 µg/ml]	[100 µg/ml]	[150 µg/ml]
<i>E. coli</i>	-	12.1±0.07	13.3±0.33	14.2±0.08
<i>P. aeruginosa</i>	-	7.1±0.20	10.9±0.24	13.4±0.32
<i>K. pneumonia</i>	-	12.5±0.47	15.1±0.38	16.4±0.26

In our results agreement with methanol extract of the *A. vulgaris* and *G. fragrantissima* was good antibacterial activity of bacterial pathogens such as *Enterococcus* sp, *Staphylococcus aureus*, *Bacillus subtilis* and *Klebsiella pneumonia* [57]. Furthermore studies reported that *S. grandiflora* extract showed antibacterial potential [58]. In addition, Different parts of plants extracts were reported to have broad biological activities such as antimicrobial, antihypertensive, antispasmodic and bronchodilator, hepatoprotective, antidepressant, xanthine oxidase inhibitor, and antioxidant [59].

### 4. CONCLUSION

The status of depression was assessed by the Sucrose consumption test which was confirmatory test of depression. Confirmatory test was done on depression induced and treated rats [Group III, IV, V, VI]. From the test, depression induced rats showed reduced sucrose solution consumption than the normal. The depression induced rats showed a drastic variation on the glucose, protein, lipid profile and aminoacids levels. The protein concentration and In the lipid profile such as Triglycerides, Total Cholesterol, and LDL also found to be low in depressed group and elevated in normal and treated groups. The Glucose, HDL, VLDL level was increased in the depressed group and reduced in normal and treated groups. When compared to normal rats, the amount of all aminoacids such

as phenylalanine, tyrosine, and tryptophan in Group II demonstrates a low amino acid activity. These levels were significantly increased when treated with antidepressant drug Imipramine, *Sesbania bispinosa* and swimming exercise. Thus it can be concluded that the plant hydroethanolic *S. bispinosa* and swimming exercise has potent antidepressant activity and proved to be a good antioxidant, antibacterial activity agent.

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