Journal of Animal Environment

(E-ISSN 2717-1396, ISSN 2717-1388)

Original Research Paper

Neuroprotective and antidepressant effects effect of *Sesamum indicum* leaf essential oil in Unpredictable chronic mild stress model of depression: Behavioral and biochemical evidences

Pratibha Rathee[1], Sandeep Kumar[2*], Tanuj Hooda[3*]

[1]M.M. College of Pharmacy, Maharishi Markandeshwar (Deemed to be University), Mullana, Haryana, India
Email id- pratibha.rathee1@gmail.com; Orchid Id: 0000-0002-6898-0514

[2]M.M. College of Pharmacy, Maharishi Markandeshwar (Deemed to be University), Mullana, Haryana, India
Email id- sranga118@gmail.com; Orchid ID: 0000-0003-3898-9582

[3] Delhi Pharmaceutical Sciences and Research University, New Delhi, India
Email id- tanujhooda2010@gmail.com; Orchid Id: 0000-0002-6701-586X

Corresponding Author Name & E-mail: Sandeep Kumar, sranga118@gmail.com; tanujhooda2010@gmail.com

Abstract

Background: *Sesamum indicum* Linn. (Pedaliaceae) is an oilseed plant with neuroprotective, antioxidant, anticonvulsant, anticancer, and wound-healing activities, primarily attributed to its lignans, phenolic acids, unsaturated fatty acids, and tocopherols.

Objective:

The present study evaluated the antidepressant potential of *Sesamum indicum* leaf essential oil and its underlying mechanisms using *in vitro* assays and a UCMS-induced rat model of depression.

Materials and Methods: GC-MS analysis was performed to identify and quantify volatile constituents of SIEO. Antidepressant activity was initially assessed by evaluating inhibition of total MAO and MAO-A. Thirty-six Wistar rats were assigned to six groups (n = 6); group I received control (p.o, unstressed control), group II-UCMS (stressed animals), group III received Fluoxetine (10 mg/kg, p.o.), and groups IV, V, and VI received SIEO-treated groups (200, 300, and 400 mg/kg, p.o). The sucrose preference consumption and tail suspension tests were used to evaluate the behavioural activity in addition to the biochemical parameters (lipoperoxidation, reduced glutathione, superoxide dismutase, catalase activity, and nitrite).

Results: GC–MS identified 69 volatile compounds in *Sesamum indicum* essential oil, with 1,3-dioxolane, 2-methyl-2-(1-methylethyl) as the major constituent. SIEO preferentially inhibited MAO-A (IC_{50}: 400 µg/mL) over MAO-T (IC_{50}: 371.43 µg/mL) and significantly alleviated UCMS-induced depressive-like behaviors and oxidative stress in rats.

Conclusion: The study revealed that both the SIEO relieved depression-like states through the mitigation of oxidative stress with a reduction in glutathione levels, superoxide dismutase and catalase activities and brain Monoamine Oxidase-A (MAO-A) levels.

Keywords: Depression, *Sesamum indicum*, Monoamine oxidase, Unpredictable Chronic Mild Stress, Sucrose preference consumption test

1. Introduction:

The majority of illness states can originate from both pathogenic and non-pathogenic factors that alter normal bodily processes. Humans mostly used bioactive substances derived from plants prior to the development of conventional medicine for the treatment and management of diseases [1]. However, people can develop depression as a result of poor disease management and therapy. Other factors that can contribute to depression include stigma, low self-esteem, and financial hardship. The development of severe depression may be influenced by genetic, hormonal, immunological, neurological, and neuroendocrine factors, although these primarily rely on how people respond to stimuli and interpret emotional data [2]. Depression is a mood condition characterized by a chronic sense of sadness and diminished interest in one's once-enjoyed activities. If left untreated, depression typically leads to suicide [3]. Some epidemiological studies indicate that about 20% of the general population will have at least one of these illnesses at some point in their lives [4]. Up to 65% of people endure recurrent periods of depression, and nearly one in four women and one in six men will experience depression at some point in

***Corresponding Author:**
Received: 25th May, 2026; Revised: 6th June, 2026; Accepted: 8th June, 2026; Available Online: 04rd August, 2026
(DOI): 10.70102/AEJ.2026.18.2.2

their lives [5-8]. Depression is now the major cause of nonfatal disease burden globally and the fourth biggest cause of overall disease burden [9]. Approximately 5% of people worldwide suffer from depression. The incidence is lower in the Western Pacific area (4.2%) and slightly higher in Africa and the Americas (5.8%) among females. On the other hand, the Western Pacific region has the lowest occurrence (2.8%), while Africa has the highest prevalence (4.8%). Additionally, onset age varies by geography; in high-income countries, it is approximately 26 years old, whereas in low- and middle-income countries, it is approximately 24 years old. By 2030, depression is expected to overtake all other causes of illness burden globally [10]. Genetic, environmental, psychological, and physiological variables are all involved in the etiology of depression. Alcoholism, childhood trauma, long-term health conditions (such diabetes, heart disease, or obesity), female sex, low socioeconomic level, advanced age, a personal or family history of depression, recent childbirth, and the occurrence of stressful life events are risk factors. To create more successful intervention techniques, it is essential to identify and address these risk factors [10]. Currently, medications including tricyclic antidepressants, monoamine oxidase inhibitors, and selective serotonin reuptake inhibitors are used to treat depression; however, they have significant side effects, including anxiety, drowsiness, weight gain, and insomnia [11]. The potential incorporation of plant-based bioactive elements and nutraceuticals may be useful in the treatment and management of depression because the majority of depressed patients are resistant to standard pharmacological administration [3]. Sesamum L. is a genus of flowering plants in the Pedaliaceae family, with 30 recognized species spread over the globe. One of the genus's most commonly grown annual plants is *Sesamum indicum*, commonly referred to as sesame [12,13]. With species found in nations including China, Malaysia, and India, it is believed to have African origins. The world's largest producers of sesame are China and India, followed by Burma, Mexico, Turkey, and Uganda. Sesame seeds have been utilized by the Chinese for almost 5,000 years, and their oil is burned to create soot for Chinese ink blocks. The phrase "OPEN SESAME," which was inspired by the Sesame seed capsule, was used in the tale of Ali Baba and the Forty Thieves, one of the Arabian Nights. Around China and India, sesame's therapeutic benefits were identified around the eighth century BC. Throughout Asia and the Middle East, whole seeds are often utilized in restaurants. In North America and Europe, bread is among the meals that use it as a flavoring and garnish [14-17]. Many phytochemical compounds, such as lignans, polyphenols, phytosterols, phenols, anthraquinones, naphthoquinones, triterpenes, cerebrosides, fatty acids, vitamins, proteins, essential amino acids, sugars, and other organic compounds, have been identified and isolated from seeds, seed oil, and different plant organs of *S. indicum* [18]. Sesame oil possesses high levels of Vitamin E, including alpha-tocopherol and gamma-tocopherol, which act as powerful antioxidants. This alleviates oxidative stress, improves skin and hair health, and promotes a youthful appearance [19]. The neuroprotective potential of the seeds, oil, and leaf extracts can be demonstrated by phenolic derivatives (sesamin, sesamol, sesaminol, sesamolin, vanillic acid, and ferulic acid), unsaturated fatty acids, and tocopherol. Consequently, it can be used as a neuroprotective drug for a range of neurological conditions [18]. Previous studies signify that Lactobacillus acidophilus and Streptococcus mutans were found to be somewhat susceptible to the sesame oil's in vitro antibacterial action against dental caries-causing bacteria. According to the study outcomes, sesame oil shows significant antibacterial activity against *S. mutans*, *Lactobacilli*, and other bacteria collectively [20]. In another study, sesame oil has been shown to successfully prevent the development of fatty acid and non-saponifiable components, perhaps leading to atherosclerotic plaques working in concert. Sesamol (INV-403) was altered to improve its qualities and evaluate its impact on atherosclerosis. A novel modified lignan derivative called Inv-403 efficiently delays the development of atherosclerosis by influencing nuclear factor-B and IKK2 signaling [21,22]. Experimental animal models have demonstrated the anti-hypertensive properties of sesamin and its active metabolites. In a study of people with hypertension, consuming sesame oil dramatically reduced oxidative stress while simultaneously raising GPx, superoxidase dismutase, and catalase activity. The researchers found that sesamin may be a useful preventative drug for cardiovascular hypertrophy and hypertension. Another study examined the use of nifedipine and other edible oils in hypertensive individuals. Sesame seems to be one of the groups that showed promise against the rise in blood pressure [22]. Moreover, The Korean Cell Line Bank (Seoul, Korea) supplied the human neuroblastoma SH-SY5Y cells. Modified Eagle's medium (DMEM) with 10% fetal bovine serum, 100 U/mL penicillin, and 50 μg/mL streptomycin was used to cultivate the cells at 37°C with 5% CO2. Phosphate-buffered saline and 0.05% trypsin-EDTA were applied to the cells. The MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyl tetrazolium bromide) test was used to measure the cytotoxicity of sesame extracts (25, 50, 100, and 200 μg/mL) and amyloid β-protein fragment 25–35 (Aβ25–35; 6.25, 12.5, 25, and 100 μM). To assess cytoprotective effects against Aβ25–35 aggregates (25 μM), SH-SY5Y cells were plated at a density of 2×104 cells/well in 96-well plates. Following a 24-hour period, cells were exposed to sesame extracts (50, 100, 200, and 400 μg/mL) for two hours before being

exposed to Aβ25–35. The MTT assay was used to evaluate cell vitality 24 hours later. A DCFH-DA fluorescent probe was used to assess the amounts of intracellular reactive oxygen species (ROS). At an excitation wavelength of 485 nm and an emission wavelength of 530 nm, fluorescence intensity was measured for two hours. Protein, lipid peroxidation, and antioxidant enzyme assays were performed on the supernatants after the cells were lysed using radioimmunoprecipitation assay buffer and centrifuged at 10,000 × g for 10 minutes at 4°C [23]. Seven *Sesamum indicum* samples, including sesame honey from Hooghly, West Bengal, India, were assessed for their polyphenol and flavonoid concentrations as well as their ability to scavenge free radicals in vitro. The polyphenol content (28.9 ± 0.6 mg GAE/100 g) and color intensity (r = 0.872–0.931) showed a good correlation with antioxidant indicators, such as IC50 [DPPH] and FRAP [ferric reducing antioxidant power]. Four flavonoids—quercetin, rutin, myricetin, and apigenin—as well as two sesame lignans (episesamin and sesamin) and a ferulic acid derivative (cinnamic acid) were found using HPLC. These results can be applied to sesame honey authentication [24]. This spice plant is thought to be crucial in the management and treatment of several stress-related illnesses, such as depression, because it is known to contain components with strong antioxidant activity. There is no recent report on the antidepressant effect of the essential oil from these spices. This study aimed to explore and compare the biochemical effects and antidepressant potentials of the essential oil from *S. indicum* leaf in experimental rats.

2. Materials and Methods

2.1 Plant materials

Leaves of the *Sesamum indicum* plant were collected in May and November of 2024, respectively, from Khairthal district, Swaroop Sarai village, Karni Kot post, Mundawar tehsil, Rajasthan, India. The plant *Sesamum indicum* was successfully identified and verified by Dr. S.S. Yadav, an associate professor in the botany department of Maharshi Dayanand University in Rohtak, Haryana.

2.2 Extraction of essential oil

The leaves of *Sesamum indicum* were thoroughly washed, dried at 35–40°C for two weeks, washed with distilled water, ground using an electric grinder, and sieved (Sieve No. 60) to remove larger particles. The resulting powder was stored in airtight containers. The coarse powder was mixed with distilled water and utilized a Clevenger device for hydro-distillation. Vapors were condensed, and the essential oil was collected in a bottle [25,26].

2.3 Analysis of essential oil constituents using GC-MS

GC-MS analysis of *Sesamum indicum* essential oil was performed using a Thermo Fisher gas chromatograph with a TSQ 8000 mass spectrometer and a BP-5MS (0.25mm x 30m x 0.25 μM). The analysis was performed using helium as the carrier gas (1 mL/min) with a 1:10 split ratio. The temperature program started at 60 °C (1 min), raised to 250 °C at 4 °C/min, and was kept for 15 minutes. Mass spectra were recorded at 70 eV. A sample was formulated by blending 30 microliters of essential oil with 970 μL of hexane, and 1 μL was injected. Identified compounds were matched with NIST 2.0 and WILEY libraries and reported as percentages of their total ion chromatogram peak areas [27].

2.4 In vitro antidepressant activity

2.4.1 Monoamine oxidase analysis

The essential oil *in vitro* monoamine oxidase (MAO) inhibitory action will be tested using the brain homogenate of untreated rats made in phosphate buffer solution (pH 7.4). The Biuret assay will be used to measure the total protein concentration in accordance with the procedure outlined by Cain et al. (1987) and standardized to a final value of 1 mg/mL.

Monoamine oxidase (MAO) inhibition will be assessed by incubating the tissue homogenate with varying concentrations of the essential oil (25, 100, 200, 400, and 800 mg/mL) or phosphate buffer as the control at 37 °C in a water bath with continuous stirring for 5 min. Subsequently, 90 mM Kynuramine, a non-selective MAO substrate, will be incorporated, and the mixture will be left to incubate for an additional 30 min under identical conditions.

To assess Monoamine oxidase-A inhibitory activity, the homogenate will first be pre-incubated with 250 nM selegiline, an MAO-B–specific inhibitory agent, for 5 min. The essential oil will then be introduced at the previously mentioned concentrations. 90 mM Kynuramine will be added after 5 minutes of incubation, and the combination will then be incubated for 30 minutes under the same conditions as before. In all experimental assays, the reactions will be stopped by adding 10% trichloroacetic acid. After centrifuging the samples, 800 mL of the supernatant will be put into new microtubes together with 1000 mL of 1 M NaOH. Three microplate wells will then be filled with 200 mL aliquots of each sample. A spectrophotofluorometer operating with excitation and emission wavelengths set at 320 nm and 460 nm, respectively, will be used to measure the concentration of 4-hydroxyquinoline, the fluorescent product produced by the MAO-mediated oxidation of kynuramine.

The inhibitory effect on total MAO activity will be expressed using the equation below:

$$\% \text{ of inhibition} = \frac{\text{Fluorescence of control} - \text{Fluorescence of sample}}{\text{fluorescence of control}} \times 100$$

For the assessment of MAO-A inhibition, the same calculation formula will be applied; however, the control fluorescence values will be substituted with the fluorescence values obtained from samples

treated with selegiline alone, without the extract. MAO-B inhibitory activity will be determined indirectly by calculating the difference between total MAO activity and MAO-A activity. Linear regression analysis of percentage inhibition as a function of final concentration, using the average data from three independent experiments, will be employed to determine the IC_{50} values for each MAO isoform [28].

2.5 *In vivo* Antidepressant activity

2.5.1 Approval of the experimental protocol

The Institutional Animal Ethics Committee (caf/mm(DU)/26/IAEC-77) reviewed and approved the study protocol for the Antidepressant activity model, the UCMS model of depression in rats, which was implemented under the ethical and husbandry standards prescribed by CPCSEA.

2.5.2 Unpredictable chronic mild stress induced Depression

Before the study began, the animals were moved from the housing facility to the experimental room in their home cages and given two to three hours to get used to their new environment. Over the course of each week, the rats were randomly assigned to experience seven distinct unexpected chronic moderate stressors; one stressor was given each day, and no stressor was repeated on consecutive days. The UCMS procedure was maintained for six weeks, while pharmacological intervention was initiated two weeks after the onset of the UCMS regimen and continued throughout the remaining study period. The study was conducted over a period of six weeks. In summary, the animals were exposed to a single stressor daily between 11:00 a.m. and 11:30 a.m., except for stress procedures extending over 24 h. The stress regimen comprised the following mild stressors:

- **Day I-** Animals were subjected to forced swimming in 37 °C water for 5 min.
- **Day II**-The animals were subjected to a 24-h wet bedding condition using a mixture of 100 mL water and 50 g litter.
- **Day III-** Twenty-four hours of feed deprivation
- **Day IV**- 90 seconds of tail pinch stimulation
- **Day V-** Water deprivation for 24 hours.
- **Day VI-** The rats underwent 5 min of forced swimming in 4 °C water and were subsequently dried using a towel.
- **Day VII**- The cages were tilted 45° from the horizontal plane and maintained in this position for 24 hours [29-31].

Table 1- Chronic unpredictable mild stress-induced depression in rats.

S.No	Model	Treatment group	No. of animals
1.	Acute toxicity study	Already reported [31,32]	…….
2.	Antidepressant activity of essential oil in UCMS-induced depression was studied individually	Acclimization (1 week) **Group I:** Control group will receive normal saline orally (1ml/kg BW) for 6 weeks. **Group II:** UCMS group (Unpredictable chronic mild stressor for 4 weeks) **Group III:** Standard drug group, diseased animals of this group will receive Fluoxetine10mg/kg p.o. for 6 week. **Group IV, V and VI:** Diseased animals will be given 200, 300 and 400 mg/kg p.o. essential oil from leaves of *S.indicum* for 6 weeks.	6 rats 6 rats 6 rats 18 rats
		Total	36 rats

2.6 Behavioural depression models

2.6.1 Sucrose preference consumption test

The sucrose preference test was conducted both prior to and following the 30-day exposure to UCMS. Along with social avoidance or isolation, anhedonia, which is defined as a decreased taste for sweet solutions, is a characteristic that is closely associated with an animal's feeling of defeat. By creating this test both before and after the UCMS, it is feasible to determine that the animals are in an anhedonic state at the conclusion of the stress phase, which is indicated by a notable reduction in their sucrose intake. Wilmer et al.'s instructions were followed for conducting the test. Rats were initially trained to experience and consume sucrose solution (1%) for a full day by being given two identical bottles at the same time: one containing sucrose solution (1%) and the other tap water. Rats were allowed to choose between the two bottles for a whole day as part of the testing protocol. In order to avoid any potential impacts of side preference on drinking behavior, the bottles were moved halfway through the testing. Weighing bottles were used to quantify the amount of water and sucrose solution consumed simultaneously by the experimental and control groups. It was determined how their initial and ultimate weights differed from one another. The

following formula was used to determine the sucrose preference [33]:

$$\% \text{ sucrose preference} = \frac{\text{Sucrose intake (g)}}{\text{Sucrose intake (g) + Water intake (g)}} \times 100$$

2.6.2 Tail suspension test

According to Can et al. (2012), the tail suspension test model of depressive-like behavior was employed to assess behavior despair, a characteristic of depression. Using an adhesive tape positioned about 1 cm from the tail tip, rats were suspended upside down on a metal rod in a tail suspension box at a height of 23.5 cm from the ground. Direct observation and video capture were used to document the whole duration of immobilization during the 6-minute test. The rat was deemed immobile when it did not attempt to grasp the sticky tape, twist its body, or jolt for a minimum of five seconds. This procedure will involve incubating 42 Albino rats with a standard and a plant essential oil [31].

Preparation of Brain Homogenate Preparation

Ice-cold saline of about 0.9% sodium chloride was used to properly clean the brain tissues before being prepared as a homogenate in chilled phosphate buffer (pH 7.4). To remove nuclear debris, the homogenized sample was centrifuged at 800 × g for 5 minutes while maintained at 4°C. Subsequently, to obtain the post-mitochondrial supernatant, the supernatant was centrifuged at 10,500 × g for 20 minutes at 4°C, which served as the sample for biochemical analyses of catalase, lipid peroxidation, nitrite, reduced glutathione, and superoxide dismutase [34].

2.7 Biochemical Parameters

1) Determination of lipid peroxidation levels

Using Wills' procedure (1965), Thiobarbituric acid-reactive chemicals were used to assess the quantity of MDA, a marker of lipid peroxidation. In summary, two hours at 37 degrees Celsius were spent incubating 0.5 milliliters of Tris and postmitochondrial supernatant, respectively. At the end of the incubation period, one milliliter of 10% trichloroacetic acid was added to the mixture, and the samples were centrifuged for ten minutes at 1,000 × g. One milliliter of the supernatant and one milliliter of 0.67% thiobarbituric acid were then boiled in boiling water for ten minutes. After chilling and adding one milliliter of double-distilled water, absorbance was measured at 532 nm. An extinction coefficient of 1.56×10^2 M^{-1} cm^{-1} was used to compute the concentration of TBARS, which was subsequently represented as milligrams of protein per nanomole of MDA. The quantity of brain MDA was expressed as nmol/mg protein using the Biuret technique to measure the protein content [34].

2) Measurement of GSH activity

Reduced glutathione (GSH) levels were measured using the Jollow et al. (1974) technique. To precipitate proteins, one milliliter of 4% sulfosalicylic acid and one milliliter of 10% post-mitochondrial supernatant were mixed together. The mixture was centrifuged at 1,200 g for 15 minutes at 4°C after being kept at that temperature for at least an hour. The assay mixture had a final volume of three millilitre and was composed of 0.1 mL of the supernatant, 2.7 mL of phosphate buffer (0.1 M, pH 7.4), and 0.2 mL of 5,5′-dithiobis-(2-nitrobenzoic acid) (DTNB; Ellman's reagent, 0.1 mM, pH 8.0). Using a molar extinction coefficient of 1.36×10^2 M^{-1} cm^{-1}, reduced glutathione concentrations were computed and expressed as µmol/mg protein. At 412 nm, the yellow tint that appeared was measured right away [34].

3) Assessment of Superoxide dismutase

The Kono (1978) method was used to quantify the superoxide dismutase effect. The assay combination was 96 µM NBT, 0.1 mM EDTA, and 50 mM Na2CO3. After transferring 2 mL of this reaction mixture to a cuvette, 0.05 mL of the supernatant from the mitochondrial separation and 0.05 mL of hydroxylamine HCl that had been pH-adjusted with NaOH were added. Over the course of two minutes, the extent of hydroxylamine auto-oxidation was determined from changes in absorbance measured at 560 nm, with readings taken every 30 and 60 seconds. The units per milligram of protein were used to express the superoxide dismutase activity [34].

4) Assessment of Catalase activity

The activity of catalase was assessed by Claiborne's method (1985). The assay system contained 3.0 mL of reaction volume, consisting of 0.05 mL of post-mitochondrial supernatant (10%), 0.019 M (H_2O_2, 1 mL), and 1.95 mL of pH 7.0 phosphate buffer (0.05 M). The rate of hydrogen peroxide breakdown was measured by tracking at 240 nm absorbance variations.

After that, the 0.07 mM H_2O_2 extinction coefficient was used to calculate catalase activity and expressed as µmol of H_2O_2 broken down per minute per milligram of protein [34].

5)Nitrite estimation

Nitric oxide production was assessed indirectly by measuring nitrite levels by the Griess reaction. 500 microliters of Griess reagent, consisting of 1% sulfanilamide in 5% phosphoric acid and 0.1% N-(1-naphthyl) ethylenediamine dihydrochloride in water, was reacted with 100 µL of post-mitochondrial supernatant for the assay. A spectrophotometer was used to measure the absorbance at 546 nm (Green et al. 1982). A sodium nitrite standard curve was used to calculate the nitrite concentration. The units of measurement for nitrite were micrograms per milliliter [34].

2.8 Analysis of Data

The data were analyzed using one–way ANOVA, followed by Bonferroni's multiple comparison tests, using GraphPad Prism 8.0.2. Results were denoted as Mean± SEM and statistical significance denoted as *, **, or *** for p<0.05,0.01 and 0.001, respectively. Values were shown with '#' representing p<0.05 when compared with UCMS.

3. RESULTS

3.1 Total yield of Essential oil

White sesame leaves essential oil obtained from *Sesamum indicum* is typically a light, pale yellow, or almost colorless oil, having a mild flavor. The yield of essential oil was calculated by the following equation:

$$y=\frac{V*100}{W}$$

$$y=\frac{450*100}{1000}$$

Yield of *S. indicum* leaves essential oil= 45%

where y is the yield of essential oil (%), V is the volume of collected essential oil (ml)
and W is the weight of the plant material (g).

3.2 GC-MS analysis

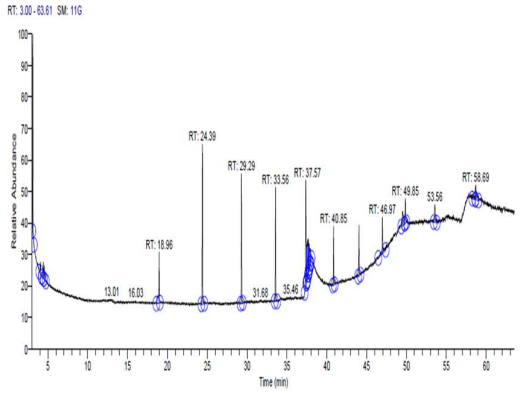


Figure 1. Peaks in the GC-MS chromatogram of *Sesamum indicum* essential oil suggest that the plant oil contains 69 different chemicals.

Table 2. List of some important chemical compounds identified in *Sesamum indicum leaves* essential oil by GC-MS results.

Compound Name	CAS	RT	Peak Area	Area %	Peak Height
2,2Dimethoxybutane	3453-99-4	3.1	33451768.1	12.93	9670914.56
1,3Dioxolane, 2methyl2(1methylethyl)	4405-16-7	3.1	33451768.1	12.93	9670914.56
2,2Dimethyl5hexen3ol	19550-89-1	3.1	33451768.1	12.93	9670914.56
tertButylpentamethyldisiloxane	67875-54-1	37.36	27758037.59	10.73	5164506.91
Disiloxane, hexamethyl	107-46-0	37.36	27758037.59	10.73	5164506.91
Ethanedioic acid, bis(trimethylsilyl) ester	18294-04-7	37.36	27758037.59	10.73	5164506.91
Ethanedioic acid, bis(trimethylsilyl) ester	18294-04-7	24.39	27141218.62	10.49	7704778.07
tertButylpentamethyldisiloxane	67875-54-1	24.39	27141218.62	10.49	7704778.07
Disiloxane, hexamethyl	107-46-0	24.39	27141218.62	10.49	7704778.07
Silane, tetramethyl	75-76-3	29.29	20425103.54	7.9	6248783.87
1,3Dioxolane, 2(3bromo5,5,5trichloro2,2dimethylpentyl	NA	29.29	20425103.54	7.9	6248783.87
1Propene1thiol	925-89-3	29.29	20425103.54	7.9	6248783.87

GC–MS analysis of *Sesamum indicum* essential oil revealed 69 volatile constituents represented in Figure 1. The predominant compounds were 1,3-dioxolane, 2-methyl-2(1-methylethyl) (12.93%), tert-butyl pentamethyldisiloxane (10.73%), ethanedioic acid bis(trimethylsilyl) ester (10.49%), and 1,3-dioxolane, 2(3-bromo-5,5,5-trichloro-2,2-dimethylpentyl) (7.9%), which together formed the major portion of the oil composition mentioned in Table 2.

3.3 In vitro antidepressant effect

3.3.1 Monoamine oxidase analysis

In our study, *Sesamum indicum* essential oil inhibited MAO, which strongly advocates the contribution of the essential oil in exerting antidepressant potential. The plant essential oil inhibited total MAO activity and the MAO-A isoform with comparable efficacy over a concentration range of 25–800 µg/mL (Figure 2), suggesting selective MAO-A inhibition at lower concentrations. However, at 800 µg/mL, inhibition of total MAO activity was markedly greater than that of MAO-A, indicating a loss of selectivity and additional inhibition of MAO-B. *Sesamum indicum* essential oil showed IC50 value for MAO-T of 371.43 µg/mL, while MAO-A was 400 µg/mL. At concentrations below 400 µg/mL, MAO-B inhibition was negligible, and at the maximum dose tested, 800

µg/mL, it was estimated to be 24%. The effect of inhibition of MAO-A was more prominent than that of MAO-B.

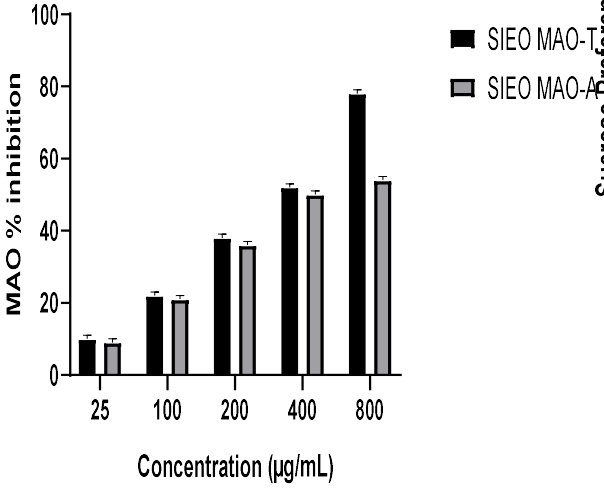


Figure 2. The inhibitory effect on monoamine oxidase (MAO) activity was expressed as percentage inhibition in rat brain homogenates. IC_{50} values were calculated through linear regression analysis of the *S. indicum* essential oil data.

3.4 In-vivo Antidepressant activity

Unpredictable chronic mild stress (UCMS) induced Depression

3.4.1 Behavioural depression models

Sucrose preference consumption test

(Figure 3) represents the percentage of sucrose preference in rats before the UCMS (A) and the effect of leaf essential oil of *S. indicum* on anhedonia (B). Before the UCMS, the sucrose preference was relatively very high (about 95%) in every animal group. After the UCMS, we noted a significant decrease in this percentage in all animals of the diseased group compared to normal animals. Nevertheless, those treated with a dose of 400 mg/kg and treated with Fluoxetine presented a nonsignificant increase in preference for sucrose compared to stressed but non-treated animals.

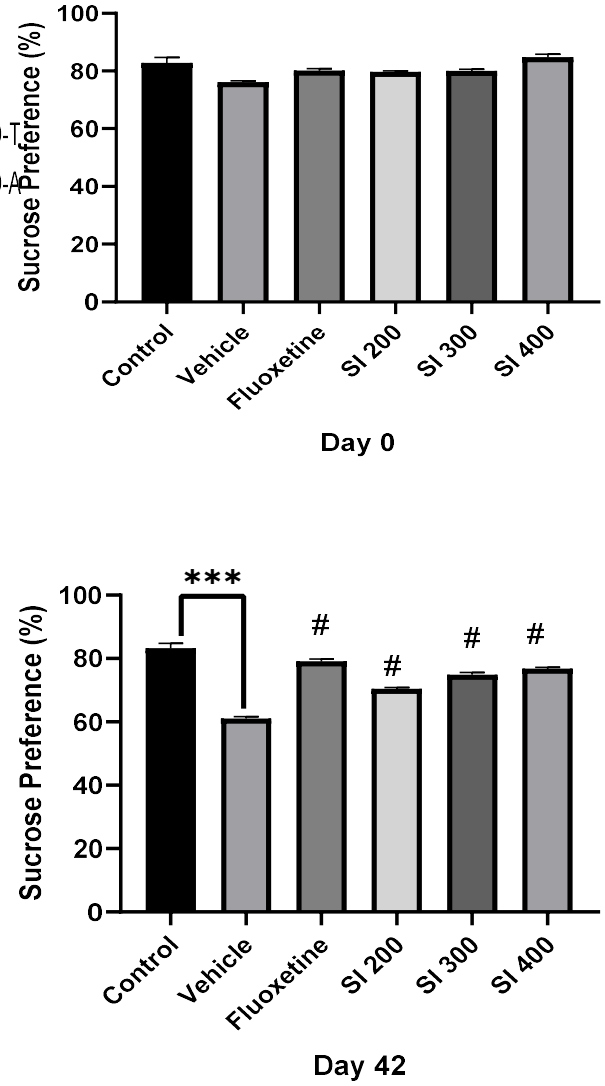


Figure 3: *Sesamum indicum* essential oil's effects on the sucrose consumption test in the UCMS rat model. The mean ± SEM (n = 6) is used to express the results. One-way ANOVA and Bonferroni's multiple comparisons test were used to assess statistical significance. NC stands for normal control; (UCMS) Unpredictable long-term mild stress; *Sesamum indicum* (SI).

Behavioral effects of essential oil in the tail suspension test

In the Tail Suspension Test (TST), rats under stress demonstrated a substantial increase in immobility time ($p < 0.001$) as compared to the normal control group. In comparison to the stress group, the period of immobility was considerably decreased by essential oil treatment where $p < 0.01$. Similarly, relative to agitated rats, fluoxetine administration significantly decreased immobility time ($p <$

0.001), suggesting an antidepressant-like effect (Figure 4).

Table 3: Immobility duration in experimental rats treated with *Sesamum indicum* essential oil against the tail suspension test.

Experimental group	Duration of immobility (sec)
Normal Control	106.1666667 ±1.301708
UCMS	187.6666667 ±1.626175
Fluoxetine (10mg/kg)	120.6666667 ± 1.229273
SIEO 200 mg/kg	158.3333333 ± 1.054093
SIEO 300 mg/kg	137.6666667 ± 0.881917
SIEO 400 mg/kg	118.8333333 ± 0.945751

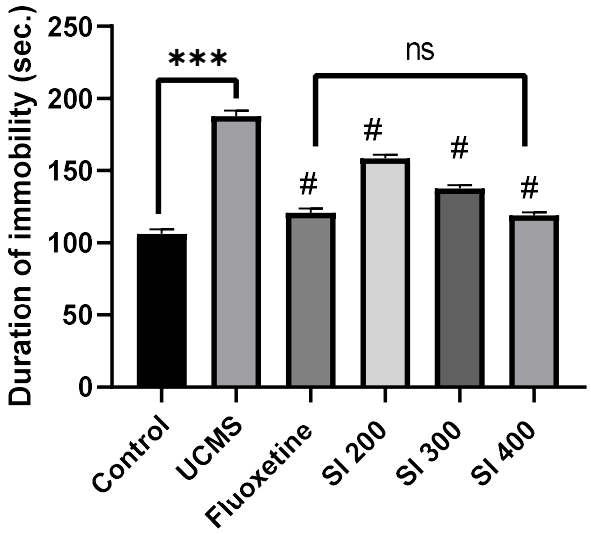
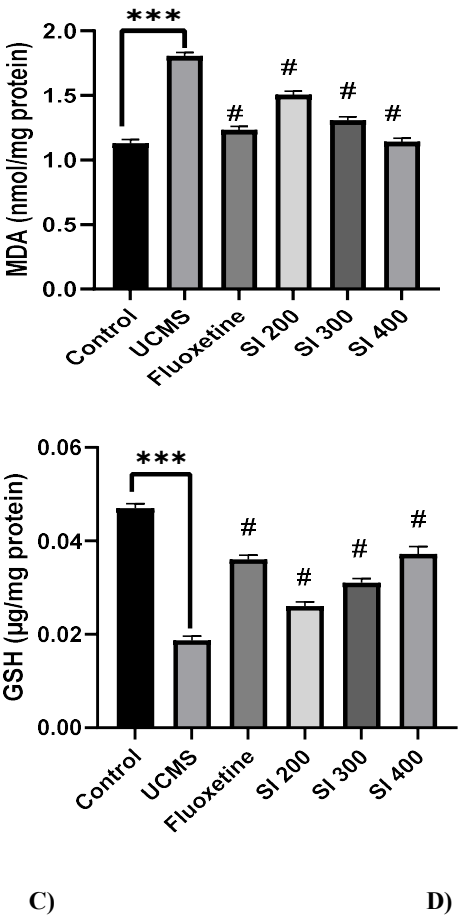


Figure 4. Evaluation of essential oil impact on tail suspension test (TST) outcomes in rats exposed to UCMS.

3.5 Biochemical Parameters



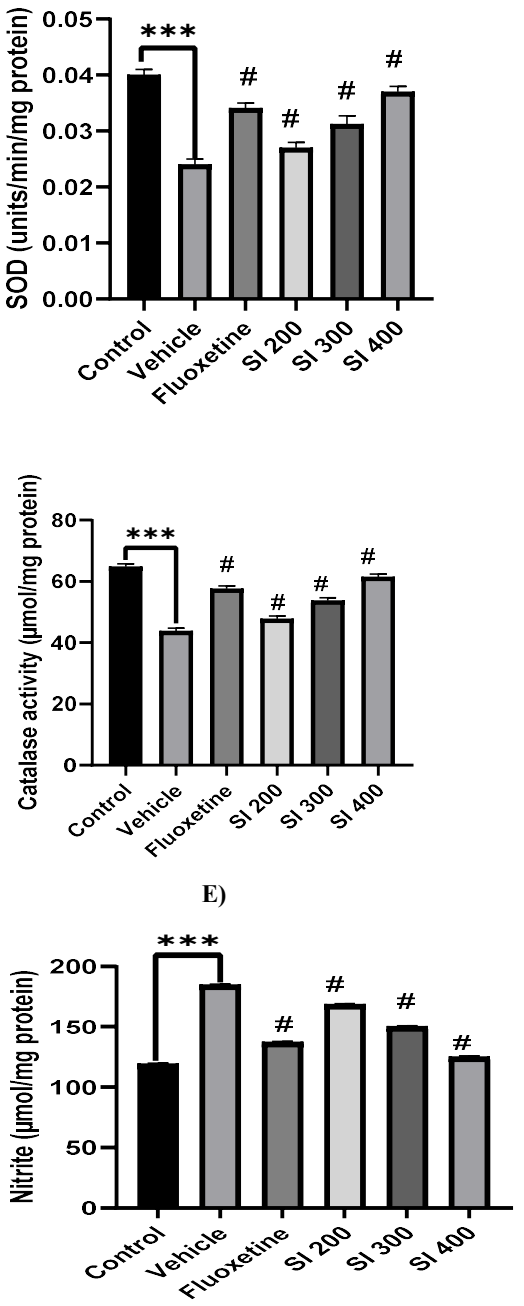


Figure 5: Effect of *S. indicum* essential oil on various biochemical parameters in UCMS-induced rats, where: A) Lipid Peroxidation, B) Reduced glutathione, C) Superoxide Dismutase, D) Catalase activity, E) Nitrite estimation

Discussion

The present study demonstrated that ***Sesamum indicum*** leaf essential oil possesses significant antidepressant and neuroprotective activity in the **Unpredictable Chronic Mild Stress (UCMS)** model of depression. The antidepressant effect was supported by phytochemical characterization, monoamine oxidase (MAO) inhibition, behavioral improvement, and attenuation of oxidative stress biomarkers. These findings suggest that the therapeutic effects of the essential oil are mediated through both **monoaminergic modulation** and **antioxidant mechanisms**, which are recognized contributors to the pathophysiology of depression. Similar multimodal mechanisms have been reported in recent experimental studies evaluating plant-derived essential oils for depressive disorders [24,25].

GC–MS analysis identified **69 volatile constituents** in *S. indicum* leaf essential oil, with 1,3-dioxolane derivatives and trimethylsilyl-containing compounds representing the predominant constituents. Although the phytochemical composition of essential oils varies depending on geographical origin, extraction method, and environmental conditions, previous investigations have emphasized that volatile phytochemicals possess antioxidant, anti-inflammatory, and neuromodulatory properties capable of influencing central nervous system function. These bioactive compounds may act synergistically to produce antidepressant effects rather than through a single active constituent [26,27].

One of the important findings of the present study was the **in vitro inhibition of MAO-A**, with comparatively less inhibition of MAO-B at lower concentrations. MAO-A is responsible for the degradation of serotonin, norepinephrine, and dopamine, neurotransmitters that are critically involved in mood regulation. Selective inhibition of MAO-A increases synaptic monoamine availability and remains an established mechanism of antidepressant drugs. The preferential inhibition of MAO-A observed in the present investigation therefore provides a plausible biochemical explanation for the antidepressant-like activity of *S. indicum* essential oil. Similar observations have been reported for several medicinal plant essential oils possessing naturally occurring MAO inhibitory compounds. [28,29]

Behavioral assessment further confirmed the antidepressant potential of the essential oil. UCMS exposure significantly reduced sucrose preference, reflecting anhedonia, which is considered one of the core symptoms of depression. Treatment with **400 mg/kg of *S. indicum* essential oil** markedly restored sucrose preference, producing effects comparable to fluoxetine. Likewise, immobility time in the Tail Suspension Test was significantly reduced following essential oil administration in a dose-dependent manner, indicating reduced behavioral despair. These findings are consistent with previous reports demonstrating that antioxidant-rich plant

extracts reverse stress-induced behavioral abnormalities through modulation of monoaminergic neurotransmission and neuroplasticity [30,31].

Chronic stress is well known to increase oxidative stress and impair endogenous antioxidant defense systems. In the present study, UCMS markedly increased lipid peroxidation and nitrite levels while significantly reducing glutathione, superoxide dismutase, and catalase activities. Administration of *S. indicum* essential oil effectively reversed these alterations, indicating substantial antioxidant and neuroprotective activity. Restoration of endogenous antioxidant enzymes may protect neuronal membranes against oxidative damage and preserve normal neurotransmitter function. Similar reductions in oxidative stress biomarkers following treatment with plant-derived essential oils have been documented in experimental depression models [32,33].

The combined behavioral and biochemical findings indicate that the antidepressant effect of *S. indicum* essential oil is likely mediated through multiple complementary mechanisms, including inhibition of MAO-A activity, reduction of oxidative stress, improvement of endogenous antioxidant defenses, and attenuation of nitrosative damage. Such multitarget activity is increasingly recognized as advantageous in managing depression, where oxidative stress, neuroinflammation, and monoamine depletion coexist. Previous experimental studies have similarly suggested that phytochemicals capable of simultaneously modulating neurotransmitter metabolism and oxidative balance exhibit superior neuroprotective potential [34].

Overall, the present findings provide experimental evidence supporting the traditional medicinal use of *Sesamum indicum* and indicate that its leaf essential oil may represent a promising natural therapeutic candidate for the management of depressive disorders. However, further investigations involving isolation of active constituents, molecular mechanism studies, pharmacokinetic evaluation, and controlled clinical trials are necessary before clinical application can be recommended.

Conclusion

The present study demonstrates that ***Sesamum indicum*** leaf essential oil exhibits significant antidepressant and neuroprotective effects in the **Unpredictable Chronic Mild Stress (UCMS)** model. GC–MS analysis identified numerous bioactive volatile constituents that may contribute synergistically to its pharmacological activity. The essential oil preferentially inhibited **monoamine oxidase-A**, improved behavioral parameters by restoring sucrose preference and reducing immobility in the tail suspension test, and effectively attenuated oxidative stress by decreasing lipid peroxidation and nitrite levels while enhancing glutathione, superoxide dismutase, and catalase activities. These findings indicate that the antidepressant action of *S. indicum* leaf essential oil is mediated through combined **monoaminergic modulation and antioxidant neuroprotection**. The study highlights the potential of *S. indicum* as a promising source of novel phytotherapeutic agents for depression. Nevertheless, additional pharmacological, toxicological, and clinical studies are required to identify the active constituents, elucidate precise molecular mechanisms, and establish its safety and efficacy in humans.

Acknowledgments
The authors are exceedingly grateful to the Head, M.M. College of Pharmacy, Maharishi Markandeshwar (deemed to be university), Ambala, Haryana, India, for providing essential amenities to accomplish this research study.

Declaration:

Ethical Approval:
This study used healthy Albino Wistar male and female rats weighing 150–200 g, with ethical approval no. (caf/mm(DU)/26/IAEC-77) from M.M. College of Pharmacy, Ambala, approved in 2026.

Competing Interest:
The authors declare no competing interests.

Authors contribution:
All authors made substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; took part in drafting the article or revising it critically for important intellectual content; agreed to submit to the current journal; gave final approval of the version to be published; and to be accountable for all aspects of the work. All the authors are eligible to be author as per the requirements/ guidelines.

Funding:
Funding for the study was done by M.M. College of Pharmacy, Maharishi Markandeshwar (Deemed to be University), Ambala, Haryana, India.

Data Availability:
All the data are available with the authors and shall be provided upon request.

References

References

1) Jiang, L. L., Gong, X., Ji, M. Y., Wang, C. C., Wang, J. H., & Li, M. H. (2020). Bioactive compounds from plant-based functional foods:

a promising choice for the prevention and management of hyperuricemia. *Foods*, *9*(8), 973. https://doi.org/10.3390/foods9080973

2) National Research Council. (2009). Committee on depression, parenting practices, and the healthy development of children. In *Depression in parents, parenting, and children: Opportunities to improve identification, treatment, and prevention*. National Academies Press (US). doi: 10.17226/12565.

3) Al-Harbi, K. S. (2012). Treatment-resistant depression: therapeutic trends, challenges, and future directions. *Patient preference and adherence*, 369-388. https://doi.org/10.2147/PPA.S29716.

4) Pedersen, C. B., Mors, O., Bertelsen, A., Waltoft, B. L., Agerbo, E., McGrath, J. J., ... & Eaton, W. W. (2014). A comprehensive nationwide study of the incidence rate and lifetime risk for treated mental disorders. *JAMA psychiatry*, *71*(5), 573-581. doi:10.1001/jamapsychiatry.2014.16

5) Kessler, R. C., Birnbaum, H., Bromet, E., Hwang, I., Sampson, N., & Shahly, V. (2010). Age differences in major depression: results from the National Comorbidity Survey Replication (NCS-R). *Psychological medicine*, *40*(2), 225-237. doi:10.1017/S0033291709990213.

6) Eaton, W. W., Shao, H., Nestadt, G., Lee, B. H., Bienvenu, O. J., & Zandi, P. (2008). Population-based study of first onset and chronicity in major depressive disorder. *Archives of general psychiatry*, *65*(5), 513-520. doi:10.1001/archpsyc.65.5.513

7) Monroe, S. M., & Harkness, K. L. (2011). Recurrence in major depression: a conceptual analysis. *Psychological review*, *118*(4), 655. DOI: 10.1037/a0025190.

8) Yiend, J., Paykel, E., Merritt, R., Lester, K., Doll, H., & Burns, T. (2009). Long term outcome of primary care depression. *Journal of affective disorders*, *118*(1-3), 79-86. https://doi.org/10.1016/j.jad.2009.01.026

9) Rahman, M. R., Sharif, M., Ali, M., & Rajib, S. S. (2019). Evaluation of the antidepressant-like activity of the methanolic extract of the seeds of Sesamum indicum. *Clinical Phytoscience*, *5*(1), 2. https://doi.org/10.1186/s40816-018-0098-3

10) Li, H. N., Gao, Y., Li, Z. K., Qiao, J. J., Peng, Y. X., Liu, S., & Yan, X. (2026). Plant-based diets and depression: epidemiological evidence, biological mechanisms, and implications for prevention. *Frontiers in Nutrition*, *13*, 1763010. 10.3389/fnut.2026.1763010

11) Zhang, Z. J. (2004). Therapeutic effects of herbal extracts and constituents in animal models of psychiatric disorders. *Life sciences*, *75*(14), 1659-1699. https://doi.org/10.1016/j.lfs.2004.04.014

12) The Plant List, 2013 Version 1.1. Published on the Internet. http://www.theplantlist. org/. (Accessed June 2021). Van Wyk, B.-E., 2006. Food Plants of the World. An Illustrate Guide

13) Morris, J. B., Wang, M. L., & Tonnis, B. D. (2021). Variability for oil, protein, lignan, tocopherol, and fatty acid concentrations in eight sesame (Sesamum indicum L.) genotypes. *Industrial Crops and Products*, *164*, 113355. https://doi.org/10.1016/j.indcrop.2021.113355

14) Amoo, S. O., Okorogbona, A. O. M., Du Plooy, C. P., & Venter, S. L. (2017). Sesamum indicum. In *Medicinal spices and vegetables from Africa* (pp. 549-579). Academic Press. https://doi.org/10.1016/B978-0-12-809286-6.00026-1

15) Anilakumar, K. R., Pal, A., Khanum, F., & Bawa, A. S. (2010). Nutritional, medicinal and industrial uses of sesame (Sesamum indicum L.) seeds-an overview. *Agriculturae Conspectus Scientificus*, *75*(4), 159-168.

16) Hegde, D. M. (2012). Sesame. In *Handbook of herbs and spices* (pp. 449-486). Woodhead Publishing. https://doi.org/10.1533/9780857095688.449

17) Mohamed, E. A., Ahmed, H. I., Zaky, H. S., & Badr, A. M. (2021). Sesame oil mitigates memory impairment, oxidative stress, and neurodegeneration in a rat model of Alzheimer's disease. A pivotal role of NF-κB/p38MAPK/BDNF/PPAR-γ pathways. *Journal of ethnopharmacology*, *267*, 113468. https://doi.org/10.1016/j.jep.2020.113468

18) Mili, A., Das, S., Nandakumar, K., & Lobo, R. (2021). A comprehensive review on Sesamum indicum L.: Botanical, ethnopharmacological, phytochemical, and pharmacological aspects. *Journal of Ethnopharmacology*, *281*, 114503. https://doi.org/10.1016/j.jep.2021.114503

19) Korde, S., Mulye, N., & Chavan, M. AN AYURVEDIC REVIEW ON TILA TAILA (SESAMUM INDICUM L).

20) Anand, T. D., Pothiraj, C., Gopinath, R. M., & Kayalvizhi, B. (2008). Effect of oil-pulling on dental caries causing bacteria. *Afr J Microbiol Res*, *2*(3), 63-66.

21) Khan, Z. J., & Khan, N. A. (2021). A COMPREHENSIVE REVIEW ON PHYTO-PHARMACOLOGICAL PROPERTIES OF SESAMUM INDICUM LINN. *International Journal of Pharmacognosy*, *8*(2), 49-57.http://dx.doi.org/10.13040/IJPSR.0975-8232.IJP.8(2).49-57

22) Hadipour, E., Emami, S. A., Tayarani-Najaran, N., & Tayarani-Najaran, Z. (2023). Effects of sesame (Sesamum indicum L.) and bioactive compounds (sesamin and sesamolin) on inflammation and atherosclerosis: A review. *Food Science & Nutrition*, *11*(7), 3729-3757.https://doi.org/10.1002/fsn3.3407

23) Kim, M. Y., Kim, S., Lee, J., Kim, J. I., Oh, E., Kim, S. W., ... & Lee, M. H. (2023). Lignan-rich sesame (Sesamum indicum L.) cultivar exhibits in vitro anti-cholinesterase activity, anti-neurotoxicity in amyloid-β induced SH-SY5Y cells, and produces an in vivo nootropic effect in scopolamine-induced memory impaired mice. *Antioxidants*, *12*(5), 1110. https://doi.org/10.3390/antiox12051110

24) Das, A., Datta, S., Mukherjee, S., Bose, S., Ghosh, S., & Dhar, P. (2015). Evaluation of

antioxidative, antibacterial and probiotic growth stimulatory activities of Sesamum indicum honey containing phenolic compounds and lignans. *LWT-Food Science and Technology*, *61*(1), 244-250. http://dx.doi.org/10.1016/j.lwt.2014.11.044

25) Kokoska, L., Havlik, J., Valterova, I., Sovova, H., Sajfrtova, M., & Jankovska, I. (2008). Comparison of chemical composition and antibacterial activity of Nigella sativa seed essential oils obtained by different extraction methods. *Journal of food protection*, *71*(12), 2475-2480. https://doi.org/10.4315/0362-028X-71.12.2475

26) Samadi, M., Abidin, Z. Z., Yunus, R., Biak, D. R. A., Yoshida, H., & Lok, E. H. (2017). Assessing the kinetic model of hydro-distillation and chemical composition of Aquilaria malaccensis leaves essential oil. *Chinese Journal of chemical engineering*, *25*(2), 216-222.http://dx.doi.org/10.1016/j.cjche.2016.09.006

27) Tyagi, R., & Sharma, V. (2014). A comparison of volatile compounds in different genotypes of Sesamum indicum L. by GC-MS. *International Journal of Pharmaceutical Sciences and Research*, *5*(1), 249-258. http://dx.doi.org/10.13040/IJPSR.0975-8232.5(1).249-58

28) Borba, L. A., Wiltenburg, V. D., Negri, G., Ibe, M. B., dos Santos, L., & Mendes, F. R. (2022). In vitro inhibition of acetylcholinesterase and monoamine oxidase by Syzygium cumini leaves extract and preliminary assessment in animal models. *South African Journal of Botany*, *146*, 553-563. https://doi.org/10.1016/j.sajb.2021.11.041

29) Ping, G., Qian, W., Song, G., & Zhaochun, S. (2014). Valsartan reverses depressive/anxiety-like behavior and induces hippocampal neurogenesis and expression of BDNF protein in unpredictable chronic mild stress mice. *Pharmacology Biochemistry and Behavior*, *124*, 5-12. https://doi.org/10.1016/j.pbb.2014.05.006

30) Singh, M. P., Vashisht, S., Chawla, V., & Mishra, P. (2016). Comparative antistress effect of Vitis vinifera and Withania somnifera using unpredictable chronic mild stress model in rats. *International Journal of Medical Research & Health Sciences*, *7*, 19-27.

31) Ekeanyanwu, R. C., Nkwocha, C. C., & Ekeanyanwu, C. L. (2021). Behavioural and biochemical indications of the antidepressant activities of essential oils from Monodora myristica (Gaertn) seed and Xylopia aethiopica (Dunal) fruit in rats. *IBRO Neuroscience Reports*, *10*, 66-74. https://doi.org/10.1016/j.ibneur.2021.01.001

32) Gholamnezhad, Z., Havakhah, S., & Boskabady, M. H. (2016). Preclinical and clinical effects of Nigella sativa and its constituent, thymoquinone: A review. *Journal of ethnopharmacology*, *190*, 372-386.

33) Foyet, H. S., Tchinda Deffo, S., Koagne Yewo, P., Antioch, I., Zingue, S., Asongalem, E. A., ... & Ciobica, A. (2017). Ficus sycomorus extract reversed behavioral impairment and brain oxidative stress induced by unpredictable chronic mild stress in rats. *BMC complementary and alternative medicine*, *17*(1), 502. https://doi.org/10.1186/s12906-017-2012-9

34) Kumar, B., Kuhad, A., & Chopra, K. (2011). Neuropsychopharmacological effect of sesamol in unpredictable chronic mild stress model of depression: behavioral and biochemical evidences. *Psychopharmacology*, *214*(4), 819-828. https://doi.org/10.1007/s00213-010-2094-2