**Antipsychotropic effects of ethanol leaf extract of Nymphaea lotus in mice**

**ABSTRACT**

Psychosis is a chronic neuropsychiatric disorder that affects millions of individuals worldwide, significantly impairing their quality of life and productivity. *Nymphaea lotus* is a plant traditionally used to manage anxiety-related disorders. This study investigates the antipsychotic effects of the ethanol leaf extract of *N. lotus* in a mouse model of ketamine-induced psychosis.

The acute toxicity of the ethanol leaf extract of *Nymphaea lotus* (ELENL) was determined using Lorke’s method. Psychosis was induced in Swiss albino mice through the administration of ketamine (25 mg/kg, intraperitoneally) for seven days. The effects of ELENL (200, 400, and 800 mg/kg, intraperitoneally) were evaluated against psychotic-like behaviors induced by ketamine. These behaviors included locomotor activity and stereotypy, measured in an open field test; immobility duration assessed in a forced swim test; and memory impairment evaluated using the Y-maze test. The apomorphine climbing test was also conducted to determine the acute antipsychotic effects of ELENL, while the woodblock test was performed to assess any extrapyramidal side effects. One-way ANOVA followed by Tukey post-hoc Multiple Comparison tests……..

The LD50 of ELENL was determined to be greater than 5000 mg/kg, indicating that it is considered safe. ELENL (200, 400, and 800 mg/kg) demonstrated significant antipsychotic effects, reducing ketamine-induced hyperactivity, immobility, and memory deficits in the mice. Additionally, ELENL suppressed stereotypic climbing behavior induced by apomorphine. Importantly, the antipsychotic activity of ELENL was not associated with extrapyramidal side effects, as evidenced by the absence of catalepsy.

In conclusion, this study demonstrates that ELENL reduced psychotic-like symptoms in mice without inducing extrapyramidal side effects, underscoring its potential as an antipsychotic agent.

Antipsychotic, *Nymphaea lotus*, Apormophine, Ketamine, Haloperidol.

1. **Introduction**

Schizophrenia is a chronic brain disorder that affects how a person thinks, feels, and behaves, often causing disruptions in thought, perception, and behavior, and can lead to hallucinations and delusions. It is a significant neuropsychiatric disorder characterized by severe and chronic mental impairment [1]. It is manifested through various symptoms that impact emotions, thoughts, perception, and will, commonly impairing the quality of life for affected individuals [2]. The symptoms of schizophrenia are generally categorized into three groups: positive symptoms (e.g., behavioural hyperactivity and hallucinations), negative symptoms (e.g., flattened affect), and cognitive symptoms (e.g., impairments in learning and memory) [3].

*Nymphaea lotus*, commonly known as the water lily, is a member of the Nymphaeaceae family. The flowers of this plant are typically white, with occasional hints of pink [4]. Native to the Nile, it is cultivated in various regions of East Africa and Southeast Asia [5]. Different tribes have their names for this plant; for example, the Igbo refer to it as ‘Ijikara,’ the Yoruba call it ‘Iyeye,’ and the Hausa know it as ‘Bado’ [6]. Water lilies primarily inhabit freshwater ecosystems, where they float on the surface of water bodies [7]. They are among the earliest aquatic macrophytes identified in Nigerian freshwater environments [8]. In traditional medicine, *N. lotus* is utilized for its various therapeutic properties, including as an aphrodisiac, anodyne, astringent, cardiotonic, sedative, demulcent, analgesic, and anti-inflammatory agent [9]. The plant contains numerous chemical compounds and has a calming effect on the nervous system, making it potentially beneficial for treating and managing insomnia and anxiety disorders [4].

Classical neuroleptics used in managing schizophrenia, such as chlorpromazine and haloperidol, are often ineffective against negative and cognitive symptoms. Moreover, the use of these conventional antipsychotics is associated with severe adverse effects, particularly extrapyramidal symptoms (EPS) and tardive dyskinesia [10]. In contrast, atypical antipsychotics have been found to alleviate positive, negative, and cognitive symptoms while reducing extrapyramidal side effects; however, they present a higher risk of metabolic disorders, such as diabetes and agranulocytosis. Additionally, long-term use of these drugs may induce oxidative imbalances, potentially worsening the progression of the disease [11]. Therefore, the search for more effective antipsychotic agents with better tolerability and fewer adverse side effects is critical. Current studies are increasingly focused on discovering new and safer molecules from natural resources to mitigate the severity and progression of mental disorders [12]. In the past decade, there has been a renewed interest in traditional medicine. This highlights the importance of exploring our indigenous plants for their potential antipsychotic properties. Thus, this study aims to investigate the antipsychotic effects of an ethanol leaves’ extract of *N. lotus* in albino mice.

1. **MATERIALS AND METHODS**
	1. **Plant Materials**

The leaves of *N. lotus* used in this experiment were collected from Iwo in Osun State, Nigeria. They were identified by a botanist in the herbarium of the Pharmacognosy Department, Faculty of Pharmacy, Obafemi Awolowo University, Ile Ife, Nigeria, where a voucher specimen was kept with the voucher number FPI 2539. The leaves were washed, air-dried, and then pulverized using a mortar and pestle.

* 1. **Extraction Procedure**

 One hundred grams of the pulverized, air-dried leaves of Nymphaea lotus were dissolved in 500 mL of distilled water in a conical flask. The mixture was shaken vigorously for 6 h and then allowed to stand for 72 h. It was subsequently filtered using Whatman (No. 1) filter paper, and the filtrate was evaporated at 50 °C in a desiccator [13].

* 1. **Anima Materials**

Healthy Swiss male albino mice weighing between 20 and 30 g were obtained from the Animal House of Ladoke Akintola University of Technology in Ogbomosho, Oyo State, Nigeria, for this study. They were maintained in a favourable environment with a temperature of 22.5 ± 2.0 °C and relative humidity ranging from 56 to 63%, following a 12-hour light/dark cycle. Their cages were cleaned daily to remove bedding and excreta. The mice were fed pelletized food sourced from Ladokun Feeds, Nigeria Limited, and had access to fresh water ad libitum during the acclimatization period. The animals were allowed to acclimate to the laboratory environment for 14 days before the experiments commenced. They underwent an overnight fasting period before the procedures. For the investigation, the mice were randomly assigned to five groups: one control group, one standard drug group, and three test groups receiving 200, 400, and 800 mg/kg. The drugs were administered via the intraperitoneal route in each group. All experimental protocols regarding animal safety and care were strictly followed.

* 1. **Drugs and chemicals**

All drugs and extracts were freshly prepared on the days of the experiments and were dissolved in normal saline. The ELENL was first dissolved in water to improve its solubility before being diluted in normal saline. The vehicle, normal saline, was administered at a volume of 10 mL per kg of body weight. The drugs used included normal saline (Dana Pharmaceuticals, Nigeria), haloperidol, apomorphine hydrochloride, risperidone, and ketamine hydrochloride (Sigma Aldrich). The chemical used was ethanol (Sigma-Aldrich, St. Louis, USA).

* 1. **Acute Oral Toxicity Study**

The acute oral toxicity of ELENL was evaluated in Swiss albino mice [14]. The study consisted of two phases. In the first phase, nine mice were divided into three groups. The mice were weighed, and the doses were calculated based on their body weight. The crude extracts were suspended in normal saline, and each group received different doses of ELENL: 10 mg/kg, 100 mg/kg, and 1000 mg/kg via oral administration.

The animals were carefully observed for signs of toxicity or mortality and monitored every 8 h for a total period of 24 h. Since no mortality was observed during this phase, the second phase of the experiment was conducted. In this phase, three mice were divided into three groups, with one mouse in each group. The groups received doses of 1500 mg/kg, 2900 mg/kg, and 5000 mg/kg, respectively, and were orally administered. The LD50 was calculated using the formula: square root of the product of D0 and D100 where D0 represents the highest dose that did not cause mortality in the mice, and D100 indicates the lowest dose that led to mortality.

* 1. **Effect of ELENL on Apomorphine-Induced Stereotypic Climbing**

The method adapted from Costall et al. [15] was used to examine the neuroleptic (anti-dopaminergic) effects of ELENL. Thirty mice were randomly assigned to five groups, with each group containing six mice. In the first group, the mice were administered 10 mL/kg of normal saline orally one hour before receiving 1.5 mg/kg of apomorphine intraperitoneally (i.p.). Risperidone, an atypical antipsychotic drug, served as the positive control in this experiment. It was administered orally at a dose of 0.5 mg/kg, also one hour before the apomorphine injection. ELENL was given intraperitoneally at doses of 200, 400, and 800 mg/kg, one hour prior to the administration of apomorphine. After the apomorphine was given, each mouse was placed in a cylindrical mesh cage, and the frequency and duration of climbing behaviour were observed for 30 min, with observations taken at 10-min intervals.

* 1. **Effect of ELENL on Spontaneous Motor Activity**

The open-field test was utilized to evaluate the effects of ELENL on spontaneous motor activity in mice. Thirty mice were divided into five treatment groups, each containing six mice (n=6). Group 1 received a normal saline solution (10 mL/kg), Group 2 received Risperidone (0.5 mg/kg, orally), and Groups 3 to 5 received ELENL at doses of 100 mg/kg, 200 mg/kg, and 400 mg/kg, respectively. One hour after administration of the treatments, each mouse was placed in the centre of an open field chamber measuring 72 cm × 72 cm × 36 cm. The number of lines crossed and the duration of the movement was recorded over five minutes [16].

* 1. **Ketamine administration**

Repeated administration of ketamine to rodents results in behavioural deficits that resemble key features of schizophrenia, including positive, negative, and cognitive symptoms [17, 18]. The effects of ELENL were examined on hyperlocomotion in the open field test, behavioural despair in the forced swim test, and spatial memory in the Y-maze test. These conditions were induced by administering daily injections of ketamine (25 mg/kg, i.p) to mice over seven consecutive days. Risperidone (0.5 mg/kg, p.o) as a positive control and ELENL (200, 400, and 800 mg/kg, i.p) were administered 1 h after each dose of ketamine [18]. The observations were noted down.

**2.9. Behavioral assessment of ELENL in Ketamine Induced Psychosis**.

2.9.1. Open-field test

 The antipsychotic-like effects of ELENL on hyperlocomotion induced by daily ketamine injections were evaluated using an open-field paradigm [17, 19]. The experiments were conducted 24 h after the final dose of a 7-day ketamine administration. Mice were individually placed in the centre square of the open field chamber, and the number of lines crossed was observed and recorded over 5 min.

 2.9.2. Y-Maze Test

The memory function was assessed through percentage alternation behaviour using the Y-maze test, as previously described by Casadesus et al. [20]. The apparatus consists of three identical arms spaced 120° apart. After a locomotor activity test conducted 24 h after the final dose of a 7-day ketamine administration, the mice were gently placed at the centre of the Y-maze and allowed to explore all three arms for 5 min. The number and sequence of arm entries were observed and recorded. The apparatus was cleaned with 70% methanol after each test session to eliminate any residual odour from the previous mice. The percentage of alternations, which measures spatial working memory was calculated by dividing the total number of successful alternations by the total number of arm entries minus one and multiplying by 100. The formula for this calculation is as follows: Percent alternations = [Total alternations / (Total arm entries - 1)] × 100.

**2.10. Effects of ELENL on Forced swim test**

The effect of ELENL on ketamine-enhanced immobility in a forced swim test was evaluated using the method described by Chindo et al. [12]. Each mouse was placed in a standardized non-opaque glass cylinder (height 50 cm, diameter 20 cm) filled with water to a depth of 30 cm at room temperature and allowed to swim for 5 min during a pretest session. Following this, mice received their treatments for 7 days, as detailed in section 2.6. One hour after the last treatment, each mouse was again placed in the same transparent glass cylinder containing water at a depth of 30 cm, and forced to swim for 6 min. The duration of immobility (i.e. the time the mouse floated in an upright position with only minor movements to avoid sinking) was recorded using a stopwatch.

**2.12. Effect of ELENL on the catalepsy test**

The effect of ELENL on catalepsy was assessed using the woodblock catalepsy test, following the modified method described by Costall and Naylor [21]. Mice were randomly assigned to 6 groups (n = 6) and were pretreated daily for 7 consecutive days with the following: normal saline, haloperidol (1 mg/kg, p.o.), ELENL (200, 400, and 800 mg/kg, i.p.), or risperidone (0.5 mg/kg, p.o.). At 60 and 90 min after the last drug administration, the mice were tested for catalepsy behaviour. Each mouse was gently held by the shoulder and below the forepaws and carefully positioned on the upper edge of a wood block surface (length = 10 cm; width = 3 cm; height = 6 cm). The descent latency was recorded as the time it took for the mouse to descend from the wood block. A mouse was considered cataleptic if it remained on the block for more than 60 seconds, as previously reported. [17].

**2.13. Statistical analysis**

The data were expressed as mean ± standard error of the mean (SEM). Statistical differences between control and treatment were determined by analysis of variance (ANOVA) with the Tukey post hoc test. The significance was considered when………????

**3. Results**

**3.1 Acute toxicity test**

The test animals did not show obvious signs of toxicity nor mortality after oral administration of 5000 mg/kg dose of ELENL. However, dullness and decreased locomotion was observed within 2 h after treatment. LD50 is greater than 5000

**Table 1a. Acute toxicity test (Phase 1I) Groups of ELENL**

|  |  |  |  |
| --- | --- | --- | --- |
| **Groups** | **Doses (mg/kg)** | **Mortality** | **% Mortality** |
| **1****2****3** | **10****100****1000** | **0/3****0/3****0/3** | **0****0****0** |

**Table 1b. Acute toxicity test (Phase 1I) Groups of ELENL**

|  |  |  |  |
| --- | --- | --- | --- |
| **Groups** | **Doses (mg/kg)** | **Mortality** | **% Mortality** |
| **1****2****3** | **1600****2900****5000** | **0/1****0/1****0/1** | **0****0****0** |

**3.2 Effects of ELENL on spontaneous motor activity (SMA) in the open-field test**

The effects of ELENL and risperidone on SMA in the open-field test are summarized in Table 1. Both the extract (800 mg/kg) and risperidone significantly reduced SMA. One-way ANOVA and Tukey post-hoc tests demonstrated that ELENL at the highest dosage of 800 mg/kg risperidone (0.5 mg/kg) significantly reduced SMA compared to vehicle-treated mice (p < 0.05) as shown in Table 2a and 2b

Table 2a: Effects of ELENL on spontaneous motor activity in open-field test

|  |  |  |
| --- | --- | --- |
| Treatments | Doses (mg/kg) | Numbers of line crossed |
| Normal salineRisperidoneELENLELENLELENL | 00.5200400800 | 105.06±6.4535.21±1.25101.59±2.3298.32±1.2530.41±0.70 |

\*\*Values are recorded as means ± SEM (n = 5).

\*Values are statistically significant (p < 0.05) in relation to control. One-way ANOVA followed by Tukey post-hoc Multiple Comparison tests.

Table 2b: Effects of ELENL on spontaneous motor activity in open-field test

|  |  |  |
| --- | --- | --- |
| Treatments | Doses (mg/kg) | Duration of ambulations (s) |
| Normal salineRisperidoneELENLELENLELENL | 00.5200400800 | 257.10±4.2398.57±1.76171.35±2.31145.32±2.39103.71±1.75 |

\*\*Values are recorded as means ± SEM (n = 5).

\*Values are statistically significant (p<0.05) in relation to control. One-way ANOVA followed by Tukey post-hoc Multiple Comparison tests.

**3.3 ELENL on ketamine-induced hyperlocomotion in mice**

The effects of ELENL and risperidone on ketamine-induced hyperlocomotion are presented in Table 3. A one-way ANOVA followed by Tukey’s post-hoc test revealed that repeated treatment with ketamine (25 mg/kg) significantly increased hyperlocomotion compared to the saline group (p < 0.05). In contrast, pretreatment with ELENL at doses of 200, 400, and 800 mg/kg (administered orally) significantly reduced hyperlocomotion induced by ketamine. Similarly, pretreatment with risperidone (0.5 mg/kg, administered orally) also significantly decreased hyperlocomotion in the ketamine-treated mice (p < 0.05).

Table 3: ELENL on ketamine-induced hyperlocomotion in mice

|  |  |  |
| --- | --- | --- |
| Treatments | Doses (mg/kg) | Numbers of line crossed\*\* |
| Normal salineKetamineRisperidoneELENLELENLELENL | 0255200400800 | 91.06±1.62135.31±1.5741.37±0.97\*89.19±3.21\*67.05±0.65\*53.37±1.42\* |

\*\*Values are recorded as means ± SEM (n = 5).

\*Values are statistically significant (p < 0.05) in comparison to control. One-way ANOVA followed by Tukey post-hoc Multiple Comparison tests.

**3.4. Effects of ELENL on ketamine-induced cognitive impairment in mice**

The effects of ELENL and risperidone on cognitive deficits induced by ketamine injection in mice, assessed using the Y-maze test are summarized in Table 4. Repeated injections of ketamine significantly reduced the percentage of alternations in performance compared to vehicle-treated mice (p < 0.05, following one-way ANOVA analysis). Pretreatment with ELENL at doses of 200, 400, and 800 mg/kg (administered orally) significantly alleviated the cognitive deficits induced by ketamine in a dose-dependent manner. Similarly, risperidone at a dose of 0.5 mg/kg (administered orally) also significantly improved memory deficits caused by ketamine treatment (p < 0.05).

Table 4: Effects of ELENL on ketamine-induced cognitive impairment in mice.

|  |  |  |
| --- | --- | --- |
| Treatments | Doses (mg/kg) | % Altermations\*\* |
| Normal salineKetamineRisperidoneELENLELENLELENL | 0250.5200400800 | 49.13±0.7325.32±0.8262.76±1.31\*47.30±1.59\*48.85±2.34\*62.01±1.97\* |

\*\*Values are recorded as means ± SEM (n = 5).

\*Values are statistically significant (p < 0.05) in relation to control. One-way ANOVA followed by Tukey post-hoc Multiple Comparison tests.

**3.5. Effect of ELENL on Ketamine-Enhanced Immobility in the Forced Swim Test**

Table 5 illustrates the effects of ELENL and risperidone on the duration of immobility induced by ketamine in the forced swim test with mice. Repeated ketamine administration resulted in a significant increase in immobility duration compared to saline-treated mice (p < 0.05). However, treatment with both ELENL (administered intraperitoneally at doses of 200, 400, and 600 mg/kg) and risperidone (0.5 mg/kg, administered orally) significantly reduced the increase in immobility duration caused by ketamine. The effectiveness of the extract in reducing immobility in mice was found to be dose-dependent.

Table 5: Effect of ELENL on Ketamine-Enhanced Immobility in the Forced Swim Test

|  |  |  |
| --- | --- | --- |
| Treatments | Doses (mg/kg) | Immobility times (s)\*\* |
| Normal salineKetamineRisperidoneELENLELENLELENL | 0250.5200400800 | 187±5.28301±4.79168±4.28\*190±1.93\*175±2.45\*168±3.15\* |

\*\*Values are recorded as means±SEM (n=5).

\*Values are statistically significant (p<0.05) in relation to control. One-way ANOVA follow by Tukey post-hoc Multiple Comparison tests.

**3.6 Effect of ELENL on Catalepsy Test in Mice**

The cataleptic effect of ELENL, as measured by the duration of immobilization in the woodblock test, is illustrated in Table 6. Mice treated with haloperidol (1 mg/kg, orally) exhibited a significant increase in duration of immobilization at the 60 and 90-minute marks compared to those treated with saline (p < 0.05). The administration of ELENL at doses of 200, 400, and 800 mg/kg via intraperitoneal injection, along with risperidone at 0.5 mg/kg, did not lead to a significant increase in the duration of immobilization at the 60-minute mark. However, at the 90-minute post-treatment interval, there was a slight increase, although it was not statistically significant when compared to the vehicle control groups.

Table 6: Effect of ELENL on Catalepsy Test in Mice

|  |  |  |
| --- | --- | --- |
| **Treatments** | **Doses (mg/kg)** | **Immobility times (s)\*\*** |
| **60-minutes** | **90-minutes** |
| Normal salineHaloperidolRisperidoneELENLELENLELENL | 010.5200400800 | 20.31 ± 1.76 67.07 ± 22.10 ± 22.50 ± 22.42 ± 23.10 ±  | 25.01± 4.17 71.21± 1.29 24. 53±1.05 24.30± 1.27 25.12± 0.86 25.40 ±0.51  |

\*\*Values are recorded as means ± SEM (n = 5).

\*Values are statistically significant (p < 0.05) in relation to control. One-way ANOVA followed by Tukey post-hoc Multiple Comparison tests.

**4.0 Discussion**

The results of this study provide evidence that the *N. lotus* leaf has potential value in managing psychotic-like symptoms. The acute toxicity study showed that *N. lotus* has an LD50 greater than 5000 mg/kg, with no observable signs of toxicity or mortality in mice. This might indicate its safety [14]. It was found to decrease the spontaneous motor activity of apomorphine-induced stereotypic climbing behaviour and ketamine-induced hyperactivity. Additionally, it demonstrated an improvement in cognitive function in the Y-maze test and decreased immobility in the forced swim test. However, haloperidol and ELENL did not inhibit the immobilization duration in the catalepsy test at 60 minutes, but it did so slightly at 90 minutes.

The ELENL dose-dependently reduced spontaneous motor activity, ketamine-induced hyperlocomotion, and apomorphine climbing behaviour, similar to the effects of risperidone. These findings suggest that ELENL possesses antipsychotic-like activity against the positive symptoms of psychosis. Previous studies have confirmed that atypical antipsychotic-like drugs alleviate symptoms such as spontaneous motor activity, ketamine-induced hyperactivity, and apomorphine-induced stereotypic climbing behaviour [17, 22]. The discovery that ELENL mitigated spontaneous motor activity, hyperactivity, and apomorphine-induced stereotypic climbing behaviour indicates its potential role in addressing psychosis. The ability of apomorphine to induce stereotypy is well characterized and documented to be mediated through the stimulation of dopamine receptors [23]. The dopamine hypothesis, a well-known theory regarding the pathophysiology of psychosis, posits that the blockade of dopamine receptors is a common mechanism of action for most antipsychotic agents [24]. Therefore, ELENL's significant attenuation of stereotyped behaviour induced by apomorphine in mice may be linked to its anti-dopaminergic activity. Ketamine-induced hyperlocomotion and stereotypy also represent aspects of the positive symptoms of schizophrenia [25], and antipsychotic agents are known to alleviate these symptoms. Furthermore, biochemical data have shown that dopaminergic and glutamatergic neurotransmitters are involved in central nervous system excitation [26]. The hyperlocomotion and stereotypic behaviours induced by ketamine are partly attributed to the blockade of NMDA receptors located on inhibitory GABAergic neurons in the mesolimbic brain regions, which leads to disinhibition and increased neuronal excitability [27]. Additionally, ketamine may act as an indirect dopamine agonist, explaining its induction of behavioural stimulation [25].

In this study, ELENL reduced the increase in locomotor activity caused by ketamine, as evidenced by a decreased number of line crossings. These findings suggest that ELENL may be a promising agent for managing schizophrenia-like hyperactivity resulting from NMDA receptor blockade within the inhibitory GABAergic system, a condition often regarded as associated with schizophrenia-related behavioural hyperactivity [28]. ELENL was found to alleviate ketamine-induced cognitive deficits in the Y-maze test in a dose-dependent manner. Similarly, risperidone also demonstrated effectiveness, as previously reported by Ben-Azu et al. [22], who noted the improvement of cognitive deficits in the Y-maze test through antipsychotic drugs. The potential of ELENL to enhance cognitive functions in psychotic disorders was evaluated using a mouse model repeatedly treated with ketamine and assessed through the Y-maze test. The Y-maze test is a well-established experimental paradigm frequently used to evaluate the memory-enhancing properties of various agents in rodents [29]. This test is based on the observation that rodents tend to remember the sequence of arm entries, a phenomenon known as spontaneous alternations. Rodents typically recall the last arm they entered to choose an alternative arm, making the Y-maze an appropriate model for assessing short-term working memory [30]. The finding that ELENL significantly mitigates cognitive dysfunction induced by ketamine in a dose-dependent manner suggests that this plant could be valuable in managing memory deficits in psychotic patients. Cognitive dysfunction is particularly relevant, given its substantial impact on the quality of life of individuals with psychosis [31].

Memory impairment caused by ketamine is associated with the antagonism of glutamatergic neurotransmission and an increase in oxidative stress in the brain. Following ketamine injection, levels of acetylcholine—a neurotransmitter that plays a crucial role in memory—are known to be reduced [18]. Additionally, an increase in acetylcholinesterase activity in the brain, particularly in the hippocampus, further supports the relationship between ketamine and memory deficits [17, 22]. ELENL also reduced the duration of immobility in the forced swim test, indicating its effectiveness against the negative symptoms of schizophrenia. Negative symptoms are a key feature of the disorder [3] and often present during the initial phase of the illness, persisting throughout its progression, even during remission periods. Increased immobility in the forced swim test—representing behavioural despair—has been previously established as a model for the negative symptoms of schizophrenia [18, 12]. The administration of ELENL resulted in decreased immobility periods in mice during this test, comparable to the effects of risperidone. Risperidone and other antipsychotic medications known for ameliorating negative symptoms typically act as antagonists of 5HT-2 receptors [References]. Therefore, this finding suggests that ELENL may possess atypical antipsychotic-like effects that warrant further investigation. The catalepsy test indicated that ELENL did not cause catalepsy in mice at the maximum dose, similar to risperidone. Catalepsy induced by typical antipsychotic agents, such as haloperidol, is attributed to the blockade of dopaminergic neurotransmission [32]. Consequently, the catalepsy test serves as a model for evaluating the propensity of antipsychotics to induce extrapyramidal side effects, which is typically performed in rodents to differentiate between typical and atypical antipsychotic agents.

**Conclusion**

The study revealed that ELENL possesses an atypical antipsychotic-like profile with modest efficacy against positive, negative, and cognitive symptoms of schizophrenia without causing extrapyramidal adverse effects. These findings suggest that ELENL is safe and may be beneficial in the treatment of psychotic-like symptoms.

**COMPETING INTERESTS DISCLAIMER:**

Authors have declared that they have no known competing financial interests OR non-financial interests OR personal relationships that could have appeared to influence the work reported in this paper.

REFERENCES

1. Joel, E. H. Lee. L., Perry B. M., Raymond W. R. and Alfred, G. G. (1996). Amphetamine Pages 219 -221. In: Goodman and Gilman The Pharmacological Basis of Therapeutics, 9th edition,

2. M.A. Geyer, F.X. Vollenweider Serotonin research: contributions to understanding psychoses Trends Pharmacol. Sci., 29 (2008), pp. 445-453, 10.1016/j.tips.2008.06.006 View articleView in ScopusGoogle Scholar

3. J.T. Coyle Glutamate and schizophrenia: beyond the dopamine hypothesis Cell. Mol. Neurobiol., 26 (2006), pp. 365-384, 10.1007/s10571-006-9062-8 Google Scholar

4. Afolayan AJ, Sharaibi OJ, Kazeem MI (2013). Phytochemical analysis and in vitro antioxidant activity of Nymphaea lotus. Int. J. pharmacol. 9(5):297-304.

5. Abu- Zaida ME, Mashaly IA, AbdEl-Monem M, Torky M (2008). Economic potentials of some aquatic plants growing in North East Nile Akinjogunla OJ, Adegoke AA, Udokang IP, Adebayo-Tayo BC (2009). Antimicrobial potential of Nymphaea lotus (Nymphaeaceae) against wound pathogens. J. Med. Plants Res. 3(3):138-141.

6. Jethro K (1994). Medicinal Plant Information. Back to Eden publishing C.O., Lonia Linda, C.A 92354, Original copy right. 11- 276. 3. J.T. Coyle Glutamate and schizophrenia: beyond the dopamine hypothesis Cell. Mol. Neurobiol., 26 (2006), pp. 365-384, 10.1007/s10571-006-9062-8 Google Scholar

7. P. Seeman Atypical antipsychotics: Mechanism of action Can. J. Psychiatry, 47 (2002), pp. 27-38, 10.1007/BF02246184 View at publisher View in ScopusGoogle Scholar

8. Akinjogunla OJ, Adegoke AA, Udokang IP, Adebayo-Tayo BC (2009). Antimicrobial potential of Nymphaea lotus (Nymphaeaceae) against wound pathogens. J. Med. Plants Res. 3(3):138-141.

9. Madhusudhanan N, Lakshmi T, Kumar SG, Venu G (2011). In vitro antioxidant and free radical scavenging activity of aqueous and ethanolic flower extract of Nymphaea Alba. Int. J. Drug. Dev. Res. 3(3): 252-258.

10. Ross, J. B. (1996). Drugs and the treatment of psychiatric disorder. Pages 399 – 430. In: Gilman and Goodman’s The Pharmacological Basis of Therapeutics.

11. Meyer, 2011 J.M. Meyer Pharmacotherapy of Psychosis and Mania L. Brunton, B. Chabner, B. Knollman (Eds.), Goodman & gilman's the pharmacological basis of therapeutics (Tenth ed.), McGraw Hill (2011), 10.1036/0071422803 Google Scholar

12. B.A. Chindo, A. Bulus, A.Y. Tijani, K.S. Gamaniel Ketamine-enhanced immobility in forced swim test: a possible animal model for the negative symptoms of schizophrenia Prog. Neuropsychopharm. Biol. Psychiatry, 38 (2012), pp. 310-316, 10.1016/j.pnpbp.2012.04.018

13 Eduardo, G., Irene, I., Emillia, C. and Angel, V. (2000). Antipsychotic effect of aqueous stem bark extract of A. andongensis in wistar albino rat. Journal o Ethnopharmacology, 70: 329 – 333.

14. D. Lorke A new approach to practical acute toxicity testing Arch. Toxicol., 53 (1983), pp. 275-287, 10.1007/bf01234480 View in ScopusGoogle Scholar

15 B. Costall, R.J. Naylor, V. Nohria Climbing behaviour induced by apomorphine in mice: a potent model for the detection of neuroleotic activity Eur. J. Pharmacol., 50 (1978), pp. 39-50, 10.1016/0014-2999(78)90251-0 View PDF View articleView in ScopusGoogle Scholar

16. R.,E. Brown, S.C. Corey, A.K. Mooren Differences in measures of exploration and fear in MHC congenic C578/61 and B6-H-2K mice Behav. Genet., 26 (1999), pp. 263-271, 10.1023/a:1021694307672 View at publisherView in ScopusGoogle Scholar

17. Lydia Doosuur Ior, Sunday Oritsetimenyin Otimenyin, Francis Kanayo Okwuasaba 2021. Antipsychotic-like effect of ethyl acetate fraction of***Terminalia macroptera***leaf in miIBRO Neuroscience Reports**.** 10: 83-89

18 M. Chatterjee, M. Verma, R. Kumari, S. Singh, A.K. Verma, A.K. Dwivedi, G. Palit Antipsychotic activity of standardized Bacopa extract against ketamine-induced experimental psychosis in mice: evidence for the involvement of dopaminergic, serotonergic, and cholinergic systems (Early Online:) Pharm. Biol., 53 (2015), pp. 1850-1860, 10.3109/13880209.2014.976350 View in ScopusGoogle Scholar

19. B.A. Ben-Azu, I.A. Omogbiya, A.M. Ajayi, E.O. Iwalewa Morin pretreatment attenuates schizophrenia-like behaviors in experimental animal models Drug Res., 67 (2017), pp. 1-9, 10.1055/s-0043-119127 View at publisherGoogle Scholar

20. V. Casadesus, K.M. Webber, C.S. Atwood, M.A. Pappolla, G. Perry, R.L. Bowen, M.A. Smith Luteinizing hormone modulates cognition and amyloid-beta deposition in Alzheimer APP transgenic mice Biochim. Biophys. Acta, 1762 (2006), pp. 447-452, 10.1016/j.bbadis.2006.01.008 View in ScopusGoogle Scholar

21. B. Costall, R.J. Naylor On Catalepsy and Catatonia and the Predictability of the Catalepsy Test for Neuroleptic Psychopharmacol (Berl), 34 (1974), pp. 233-241, 10.1007/BF00421964 View in ScopusGoogle Scholar

22. B. Ben-Azu, A.O. Aderibigbe, I.A. Omogbiya, A.M. Ajayi, O. Owoeye, T. Olonode, E.O. Iwalewa Probable mechanisms involved in the antipsychotic-like activity of morin in mice Biomed. Pharmacother., 105 (2018), pp. 1079-1090, 10.1016/j.biopha.2018.06.057

23. V. Pandy, M. Narasingam, Z. Mohamed Antipsychotic-like activity of noni (Morinda citrifolia Linn.) in mice BMC Complement Altern Med., 12 (12) (2012), p. 186, 10.1186/1472-6882-12-186 PMID: 23082808; PMCID: PMC3487797" View in ScopusGoogle Scholar

24. Beekman, A. T. F., Deeg, D. J. H. and Braam A. N. (1999). Consequences of major and minor depression in later life. A study of disability, well-being and service utilization. Psychological Medicine, 27: 307 – 311.

25. Tripathi, K. D. (2003). Drugs used in mental illness: Antipsychotic and anti-anxiety drugs. Pages 390 – 404. In: Essentials of Medical Pharmacology, 5th edition. JP Brothers Medical Publishers (P) Limited, New Delhi India.

26. M. Yadav, M. Parle, N. Sharma, D.K. Jindala, A. Bhidhasra, M.S. Dhingra, S. Dhingra Protective effects of Spinacia oleracea seeds extract in an experimental model of schizophrenia: possible behavior, biochemical, neurochemical and cellular alterations Biomed. Pharmacother., 105 (2018), pp. 1015-1025, 10.1007/s10787-017-0366-8 View articleView in ScopusGoogle Scholar

27. S.M. Stahl Beyond the dopamine hypothesis to the NMDA glutamate receptor hypofunction hypothesis of schizophrenia CNS Spectr., 12 (2007), pp. 265-268, 10.1017/S1092852900021015 View in ScopusGoogle Scholar

28. Lees, P. (1991). Central nervous system transmitters and mechanism of action of centrally active drugs. Pages 315 – 327. In: BRANDER, G. C., PUH, D. M., BYWATER, R. J. and JENKINS, W. L (Eds.) Veterinary Applied Pharmacology and Therapeutics. 5th Edition, Bailliere Tindal, London.

29. G.R. Dawson, C.M. Heyes, S.D. Iversen Pharmacological mechanisms and animal models of cognition Behav. Pharmacol., 3 (1992), pp. 286-297

30 V. Casadesus, K.M. Webber, C.S. Atwood, M.A. Pappolla, G. Perry, R.L. Bowen, M.A. Smith

Luteinizing hormone modulates cognition and amyloid-beta deposition in Alzheimer APP transgenic mice

31. Ayd. F. J. and Blackwell, B. (1970). Discoveries inbiological psychiatric. J. B. Lippincott Company, Philadelphia.

32. Mark, A. G. and Athina, M. (2000). Animal models for psychiatric disorders. Pages 231 – 234. In: Psychopharmacology, 4th Edition, Blackwell Science Limited, United Kingdom