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A Promising Strategy for Treating Alzheimer's Disease with *Origanum majorana extraction*

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ABSTRACT

Background: Alzheimer's disease is a neurodegenerative condition that causes a gradual decline in memory and cognitive function resulting from a notable decline in cholinergic neurons in certain regions of the brain. In this study dried leaves extracted from *Origanum* majorana are tested by HPLC to determine the chemical composition and phenolic compounds.

Objective: This study aimed to evaluate the potential neuroprotective effects of Origanum majorana extract in mitigating Alzheimer's disease (AD) symptoms induced by aluminum chloride ($AlCl_3$) in an experimental rat model.

Materials and methods: High-grade aluminum chloride ($AlCl_3$) and Origanum majorana leaves were used, with plant verification and chemical profiling. Male rats (n=30) were divided into five groups, receiving $AlCl_3$ to induce AD, followed by different doses of O. majorana for 30 days. Hematological, biochemical, and histopathological assessments were performed on blood, brain, and liver samples.

Results: The dried leaves of *Origanum majorana* had the largest percentage of carbohydrates (58.58%), followed by moisture, dietary fiber, protein, fat, and ash (25.72%, 15.54%, 4.52%, 1.71%, and 3.98% respectively). Besides, *Origanum majorana* leaves have high contents of terpenoids, high amounts of steroids and flavonoids, moderate amounts of coumarin, saponins, and alkaloids. Querectin, Vanillin, Coumaric acid, Naringenin, Ellagic acid, Cinnamic acid, and Propyl Gallate may have contributed to the polyphenol levels and their potential for therapeutic use. Significant decreases in lymphocyte ratio and count values were seen, along with notable variations in contrast with the control group.

Conclusion: Origanum *majorana* exhibited neuroprotective potential against Alzheimer's disease (AD) through its rich bioactive profile, including flavonoids, polyphenols, and alkaloids, which contribute to anti-inflammatory, antioxidant, and anticholinesterase activities. Significant variations in inflammatory biomarkers and lymphocyte counts suggest its influence on immune responses.

Keywords: *Origanum majorana,* Acetylcholine esterase (AChE), Alzheimer's disease (AD).

INTRODUCTION

Due to its complex origin and lack of disease-modifying therapies, Alzheimer disease (AD), which affects 60% to 70% of cases of dementia, is thought to be the most common type and has been identified as a worldwide health concern [(1)].

Alignment, speech, language, cognitive function, and decision-making abilities are all gradually diminished by this. Neurodegeneration due to microglial cell proliferation, astrogliosis, and the accumulation of β-amyloid (Aβ) peptides, commonly known as amyloid or senile plaques. While some of the symptoms of dementia can be treated, there is currently no known cure for the condition. However, there are ways to slow down the disease's growth. Increased Aβ peptide sequences have been found in AD investigations throughout the past few decades. These sequences are responsible for generating neuronal inflammation and neuroapoptosis, which eventually leads to cognitive impairment and AD [(2-3)].

Amyloid precursor protein (APP) is divided by β and γ enzyme, which then secretes a transmembrane glycoprotein to create the β-amyloid peptide. Therefore, through lowering excitatory impulses at the synapse, generating memory, neuroapoptosis, attenuating synaptic function, diminishing spinal density, and causing spinal injury, Aβ plays a crucial role in the etiology of AD and has a major impact[(4)]. The study conducted a valuable molecular basis for the diagnosis and treatment of AD has been established by research on Aβ peptides [(5)].

Changes have occurred in the kidney and bladder, including decreased kidney size, decreased creatinine clearance, nephron loss, decreased susceptibility to sodium-potassium loading, and decreased glomerular function [(6)]. Research has shown differences in biochemical properties, such as severe oxidative stress, mitochondrial failure, and cell cycle malfunction, in the lymphocytes of patients with AD and mild cognitive impairment (MCI). These observations point to a tight relationship between anemia and AD. Additionally, it has been noted that altered levels of triglycerides (TGA), total cholesterol (TC), vitamin D, vitamin B12, high-density lipoprotein (HDL), low-density lipoprotein (LDL), and glycosylated hemoglobin [(7)], have been linked to AD, as well as blood platelets with high levels of (APP) expression [(8)]. The onset and course of dementia are impacted by these alterations.

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Origanum *majorana* (OM) is an evergreen subshrub that is perennial and typically found in the Mediterranean region. This herb presents a strong and pleasant fragrance and had been used as a spice. OM is used extensively in folk medicine as a cure for a number of illnesses, including rheumatism, headaches, dyspepsia, and asthma [(9)]. OM has a high concentration of flavonoids and phenolics [(10)], which have potential pharmacological properties including antioxidant activities and acetylcholinesterase inhibition [(11-12)].

This study aimed to evaluate the potential neuroprotective effects of *Origanum majorana* extract in mitigating Alzheimer's disease (AD) symptoms induced by aluminum chloride ($AlCl_3$) in an experimental rat model. By analyzing the plant's phytochemical composition using HPLC and assessing its antioxidant, anti-inflammatory, and anticholinesterase properties, the study aimed to determine whether *O. majorana* can modulate biochemical, hematological, and histopathological markers associated with AD. The findings will contribute to the understanding of *O. majorana* as a promising natural therapeutic agent for neurodegenerative disorders, particularly in improving cognitive function and reducing neuroinflammation.

MATERIALS AND METHODS

Drugs and chemicals: Sigma Aldrich Chemical Co., located in St. Louis, Missouri, in the United States, was the supplier of aluminum chloride ($AlCl_3$) hydrate (cat. # 229393). For this experiment, the best analytical grade of chemicals and reagents were used.

Plant material: Dried leaves of *Origanum majorana* were collected from the Nursery of Agriculture Faculty, Zagazig University, Egypt. Verification of the plant material was conducted at the Department of Botany, Faculty of Science, Mansoura University, Egypt, following approved standards. For future reference, a sample voucher (Voucher number: 17985) was deposited.

Chemical composition: The moisture, protein, fat, ash, and crude fibre contents of dried *Origanum majorana* leaves were examined, as well as the chemical composition of the plant (g/100g). Total carbohydrates were calculated according to **Paudel *et al.*** [(13)].

Phytochemical screening: The conventional Harborne protocols [(14)] were used for phytochemical screening in order to identify flavonoids, alkaloids, tannins, saponins, and terpenoids.

HPLC analysis of phenolic compounds: A set of Agilent 1260s was used to perform the HPLC analysis. Using an Eclipse Plus C18 column (4.6 mm x 250 mm i.d., 5 μm), the separation was carried out. At a flow rate of 1 milliliter per minute, the mobile phase was composed of water (A) and 0.02% tri-floro-acetic acid in acetonitrile (B).

Experimental animals: Thirty male rats of average weight 100 ± 20 g were housed in five clear plastic cages with consistent free access to food and water light/dark cycles. The National Cancer Institute at Cairo University in Egypt provided the animals. The animal studies followed the ARRIVE criteria that were supplied by Zagazig University's institutional animal care and use committee **(ZU-IACUC/1/F/222/2023)** in Egypt.

Experimental design:

After the acclimatization phase, the rats (**n** = 30) were weighed and split into five equal groups, each containing six rats. The rats were subsequently subjected to the following regimen for a period of seven weeks. $AlCl_3$ solution (1600 ppm in distilled water) was fed to rats. Aluminum chloride was taken orally (0.5 ml/100 g of body weight) every day for two weeks. This dosing regimen for aluminum chloride was selected due to its low death rate and high rate of induction, as per prior publications [(15)].

a) Group I (Negative control): Normal control that received saline orally.
b) Group II (Positive control): This group is called with (AD) group, which received $AlCl_3$ dissolved in dist. Water (0.5 ml/100 g body weight) to induce the AD model for 2 weeks.
c) Group III: Received $AlCl_3$ (2 weeks) + 0.25 ml of *Origanum majorana* orally administered for 30 days.
d) Group IV: Received $AlCl_3$ (2 weeks) + 0.5 ml of *Origanum majorana* orally administered for 30 days.
e) Group V: Received $AlCl_3$ (2 weeks) + 1 ml of *Origanum majorana* orally administered for 30 days.

Hematological measurements and biochemical analysis: The blood samples were collected for hematological and biochemical analyses. The blood samples were analyzed for a number of parameters, including the total leucocytic count (WBCs), platelet count, hemoglobin (Hb) concentration, packed cell volume (PCV), mean cell volume (MCV), mean cell Hb (MCH), mean cell Hb concentration (MCHC), neutrophil, lymphocyte, monocyte, Eosinophil, and basophil counts, as well as the complete blood count (CBC), which counts the number of red blood cells (RBCs). Additionally, acetyl cholinesterase, triglycerides (TGA), total cholesterol (TC), low-density lipoproteins (LDL), high-density lipoproteins (HDL), and triglycerides (LDL) were analyzed.

Histopathology studies of rat brain and liver: Crucial organs, including the liver and brain, were removed and preserved in 10% formalin solution. Sections were cut to a thickness of 3 μm, pathologically inspected with an

Olympus BX51 microscope equipped with a digital camera and stained with hematoxylin and eosin (H & E).

Ethical approval: The Institutional Animal Care and Use Committee (ZU-IACUC/1/F/222/2023) of Zagazig University's Faculty of Medicine in Egypt approved the study. The Faculty of Medicine at Zagazig University, Egypt's Institute of Laboratory Animal Resources authorised all experimental protocols including the care of the experimental mice.

Statistical analysis

The average ± standard error of mean (SEM) obtained from every experiment in the corresponding group was used to represent the data. $P \leq 0.05$ was considered significant for all data analysis. One-way analysis of variance (ANOVA) and the SPSS version 25 software program (Windows operating system) were used to establish the statistical significance values. Turkey's multiple comparison test was used for individual comparisons.

RESULTS

Chemical composition of dried leaves of *Origanum majorana*: This study examined the moisture, total carbohydrates, crude fibre, ash, protein, and fat content of dried *Origanum majorana* leaves.

The findings indicated that the dried leaves of *Origanum majorana* had the largest percentage of carbohydrates (58.58%). This was followed by the following percentages: Moisture, dietary fiber, protein, fat, and ash (25.72, 15.54, 4.52, 1.71, and 3.98 respectively). The findings listed in table (1) showed low fat content (1.71). Because they increase Aβ peptide accumulation and other neurodegenerative indicators in AD, unhealthy eating habits, such as a Western diet or a high-fat, high-glycemic and high-cholesterol diet, are important risk factors for neurodegenerative diseases.

Table (1): Chemical composition of dried leaves of *Origanum majorana* (g/100g).

Nutrients	Amounts (%)
Total carbohydrate	41.53
Moisture	25.72
dietary fiber	15.54
Protein	11.52
Fat	1.71
Ash	3.98

Phytochemical screening: As shown in table (2), the phytochemical screening of an aqueous extract of *Origanum majorana* leaves using the specified experiments revealed the presence of high contents of terpenoids, excess amounts of steroids and flavonoids, measured amounts of coumarin and saponins, and moderate amounts of alkaloids.

Table (2): Phytochemical screening of aqueous extract of dried leaves of *Origanum majorana*

Test	Result
Terpenoids	++++
Steroids	++++
Flavonoid	++++
Coumarine	+++
Saponins	+++
Alkaloids	++

Highly present ' + + + +', moderate present ' +++ ', present ' ++ '

Total phenolic compounds: According to HPLC analysis, the compounds shown in Figure (1): Catechin, Chlorogenic acid, Querectin, Coumaric acid, Vanillin, Naringenin, Ellagic acid, Propyl Gallate, and Cinnamic acid. All these may have contributed to the polyphenol levels and their potential for therapeutic use.

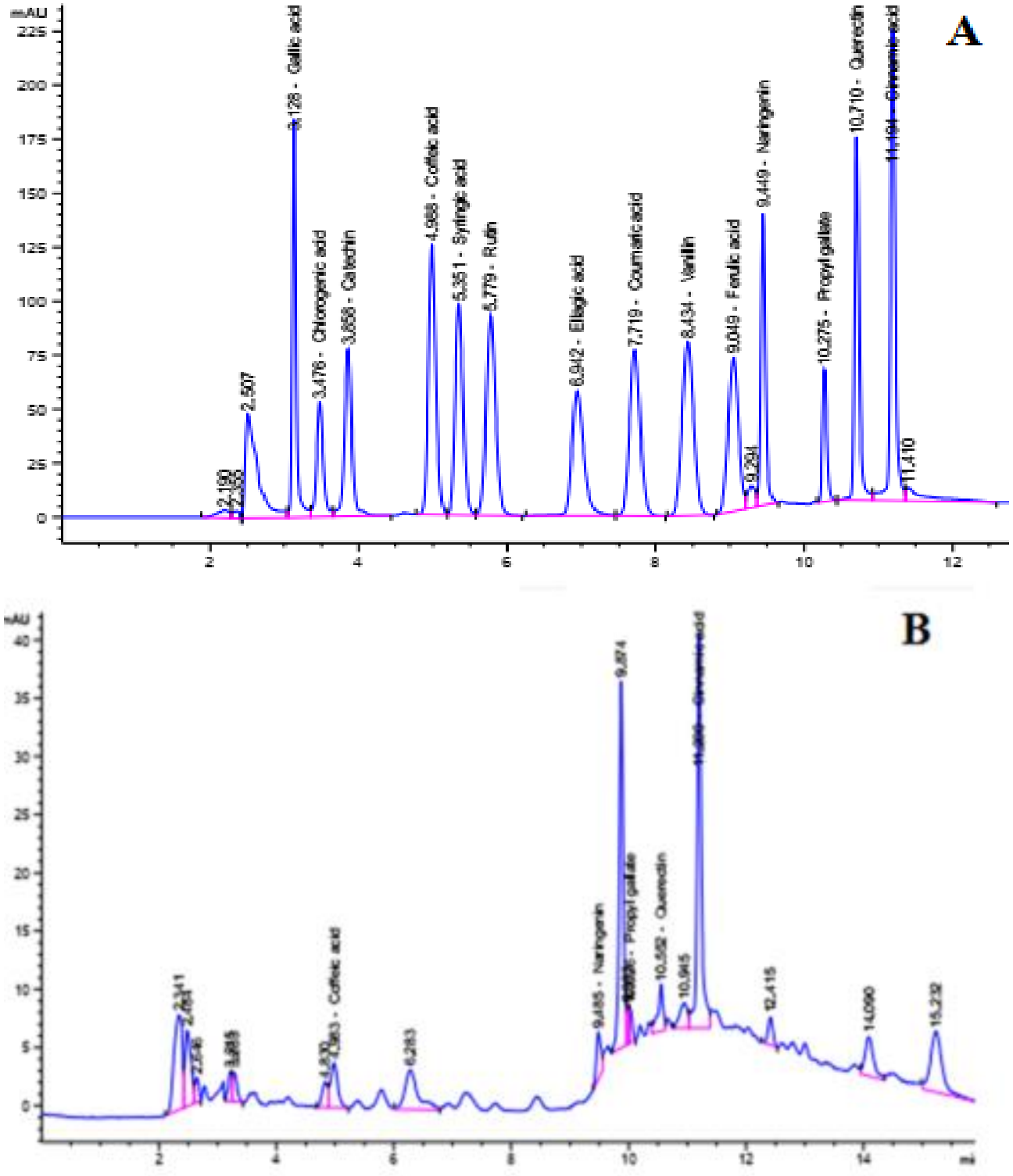

Figure (1): HPLC chromatogram: A) Standard mixture of polyphenolic compounds; B) ethanolic extract of dried leaves of *Origanum majorana*.

Histopathological examination: To investigate pathological alterations in the rat liver, histopathological analysis was performed, as seen in figure (2). (A1) A central vein (Cv) containing hepatocytes (H) arranged in usual cord configuration. These hepatocytes are characterized by their pale acidophilic cytoplasm and rounded, vesicular nuclei. One can see some bi-nucleated hepatocytes (circle) and radiating regular sinusoids (s). (B1) A single bile duct and a normal-appearing portal vein (Pv) (Bd). Observations reveal hepatocytes (H) with rounded vesicular nuclei and pale acidophilic cytoplasm. It is possible to see some bi-nucleated hepatocytes (circles). (A2) An uneven hepatocyte (H) arrangement paired with an irregular central vein (Cv). The majority of hepatocytes (H) have acidophilic cytoplasm and tiny, spherical nuclei. The nuclei of the other hepatocytes (h) are darkly pigmented, and the cytoplasm is acidophilic. A small number of bi-nucleated hepatocytes and irregularly dilated, crowded sinusoids**.** (B2) Multiple bile ducts (Bd), a clogged dilated portal vein (Pv), and an infiltration of inflammatory cells (IF). Hepatocytes (H) have pale or vacuolated cytoplasm and rounded nuclei. There are no many bi-nucleated hepatocytes (circles). (A3) An atypical hepatocyte (H) arrangement in the central vein (Cv). The majority of hepatocytes (H) have small, spherical nuclei and acidophilic cytoplasm. The cytoplasm of the other hepatocytes (h) is acidophilic, and their nuclei are darkly colored.

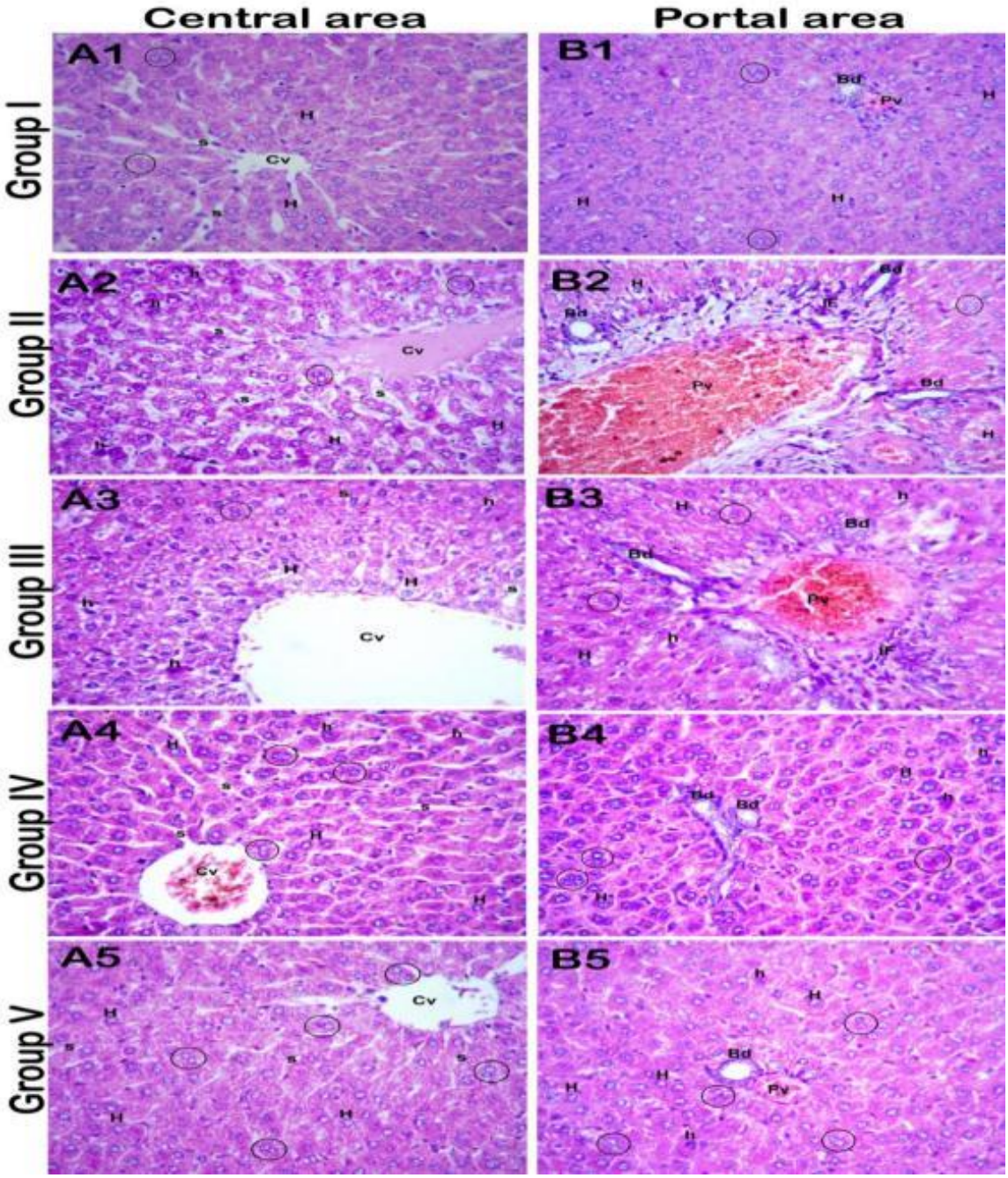

Figure (2): Microphotographs of liver hepatocyte from the different studied groups; group I, A1, B1; group II A2, B2, group III A3, B3, group IV A4, B4 and group V A5, B5 (A for central area & B for portal area), Haematoxylin-eosin staining (H&E) x400.

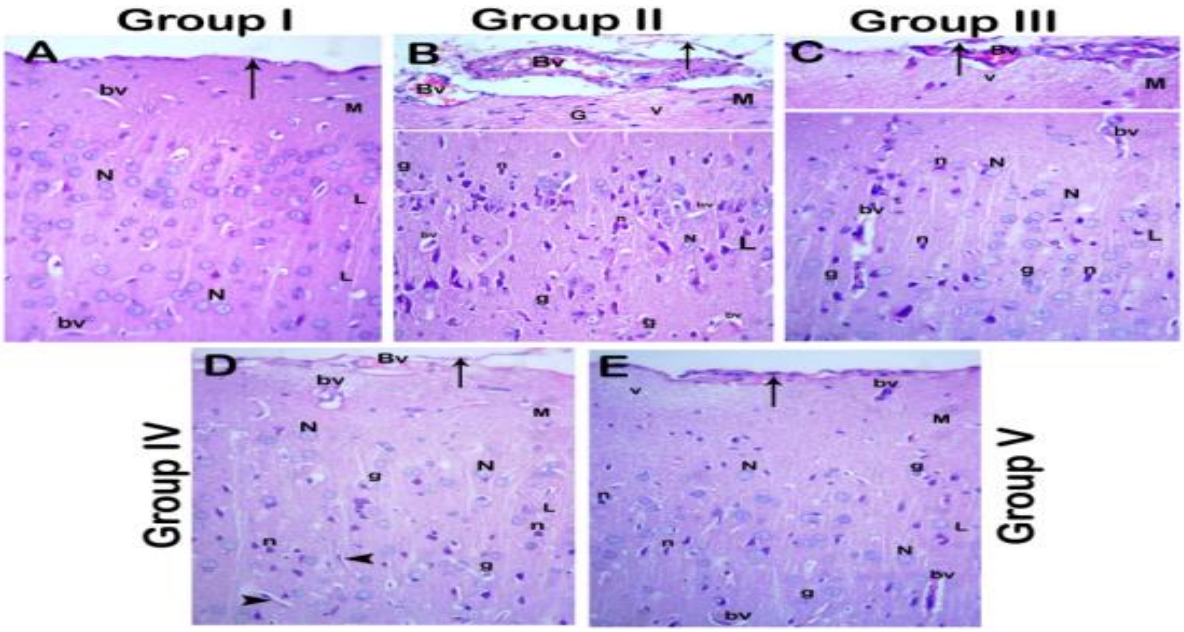

Figure (3): Microphotographs of cerebral cortex from the different studied groups; group I, A; group II, B; group III, C; group IV, D and group V, E. Haematoxylin-eosin staining (H&E) x400.

Histopathological analysis of the cerebral cortex of rats, as depicted in figure (3). A: Normal meningeal layer (arrow) and blood capillaries in the exterior cortical layers (L) and molecular layer (M) with small perivascular spaces (bv). It is possible to observe cortical neurons with basophilic cytoplasm (N) and vesicular nuclei. B: There are many glial cells (G), dilated, clogged blood vessels (Bv), and a discernible separation of the meningeal layer (arrow).

Table (3): The results of hematological measurements in all groups

Parameters	Group I	Group II	Group III	Group IV	Group V
Red Blood Cells (RBCs) (/mm^3) Count	8.22x10^6 ± 0.474	9.02x10^6 ± 0.464	8.22x10^6 ± 0.474	6.98x10^6 ± 0.338	7.65x10^6 ± 0.402
Haemoglobin (Hb) Concentration (g/dl)	15.7± 0.674	15.3± 0.607	11± 0.554	11.9± 0.574	13.7± 0.599
Packed Cell Volume (PCV) (%)	46.8± 1.502	48.2± 1.556	39.7± 1.452	35.1± 1.402	42.5± 1.489
Mean Cell Volume (MCV) (μ^3)	63.3± 2.387	59.6± 2.145	54.1± 2.136	57.4± 2.258	61.9± 2.306
Mean Cell Hb (MCH) (pg)	20.5± 0.895	18.4± 0.797	14.8± 0.785	18.3± 0.795	19.3± 0.883
Mean Cell Hb (g/dl) Concentration (MCHC)	29.1± 0.995	28.7± 0.975	25.3± 0.865	29.5± 0.999	28.4± 0.984
RBCs Distribution Width (RDW) (mcL)	13.9± 0.597	17.8± 0.697	27.4± 0.897	23.1± 0.799	19.6± 0.897
Platelet Count (mcL)	755x10^3 ± 69.571	570x10^3 ± 54.751	755x10^3 ± 69.571	720x10^3 ± 69.501	905x10^3 ± 75.345
Total Leucocytic Count (WBCs) (mcL)	18.2x10^3 ± 0.790	16.3x10^3 ± 0.609	9.6x10^3 ± 0.495	17.3x10^3 ± 0.708	18.3x10^3 ± 0.795
Neutrophils (x10^9/L)	3.28x10^3 ± 0.045	3.42x10^3 ± 0.052	4.51x10^3 ± 0.063	3.46x10^3 ± 0.050	5.31x10^3 ± 0.075
Lymphocytes (x10^9/L)	14.01x10^3 ± 0.621	11.74x10^3 ± 0.565	4.42x10^3 ± 0.060	12.98x10^3 ± 0.572	11.71x10^3 ± 0.550
Monocytes (x10^9/L)	0.73x10^3 ± 0.001	0.98x10^3 ± 0.019	0.48x10^3 ± 0.001	0.69x10^3 ± 0.032	0.92x10^3 ± 0.019
Eosinophils (%)	0.18x10^3 ± 0.001	0.16x10^3 ± 0.001	0.19x10^3 ± 0.001	0.17x10^3 ± 0.001	0.37x10^3 ± 0.001
Basophils (x10^9/L)	0.00	0.00	0.00	0.00	0.00

Table (4) showed that rats with $AlCl_3$-induced AD had significantly higher levels of AChE-RBC than the control group, which may indicate that these rats' neurotransmission abilities are severely impaired.

Table (4): The results of the biochemical analysis of blood in all groups

Parameters	Group I	Group II	Group III	Group IV	Group V
Total cholesterol(mg/dL)	108± 4.40	89± 3.99	92± 4.04	87± 3.97	79± 3.39
HDL(mg/dL)	45± 2.56	36± 2.23	36± 2.23	35± 2.23	33± 2.21
LDL(mg/dL)	51± 3.03	39± 2.26	38± 2.26	38± 2.26	35± 2.23
VLDL(mg/dL)	12± 0.57	14± 0.620	18± 0.790	14± 0.620	11± 0.54
Triglycerides(mg/dL)	67± 3.28	74± 3.35	89± 3.99	71± 3.31	55± 3.10
Acetyl cholinesterase(U/L)	5.1± 0.07	9.4±0.04	11.3± 0.55	6.2± 0.08	2.7± 0.03

The rat CA1 region of the hippocampal region underwent histopathological analysis to investigate pathological alterations. The rat hippocampus of the control group was photographed with H & E x40 stained sections to reveal distinct portions of the hippocampal development. The hippocampus proper is made up of the Cornu Ammonis (CA) regions CA1, CA2, CA3, and CA4. CA4's upper and lower limbs are surrounded by the dentate gyrus (DG). **B:** Its three levels are distinguishable: the polymorphic layer (PL), the molecular layer (M), and the pyramidal layer (P). The pyramidal layer consists of three to five dense layers of pyramidal neurons, the majority of which had vesicular nuclei (N). It should be noted that the blood capillaries and glial cells (g) with thin perivascular gaps (bv) were present in both the molecular and polymorphic layers. **C:** Three layers are observed: the pyramidal layer (P), the polymorphic layer (PL), and the molecular layer (M). Layers of pyramidal neurons are displayed, the majority of which have vesicular nuclei (N). There are some shrinking neurons with pericellular haloes (n) and darkly stained nuclei, as well as some cell loss (asterisk). There are many blood capillaries and glial cells (g) with large perivascular gaps (bv) in both the molecular and polymorphic layers. **D:** Its three layers are visible: the polymorphic layer (PL), the pyramidal layer (P), and the molecular layer (M). In the pyramidal layer, the majority of the pyramidal neurons are shown as having vesicular nuclei (N).

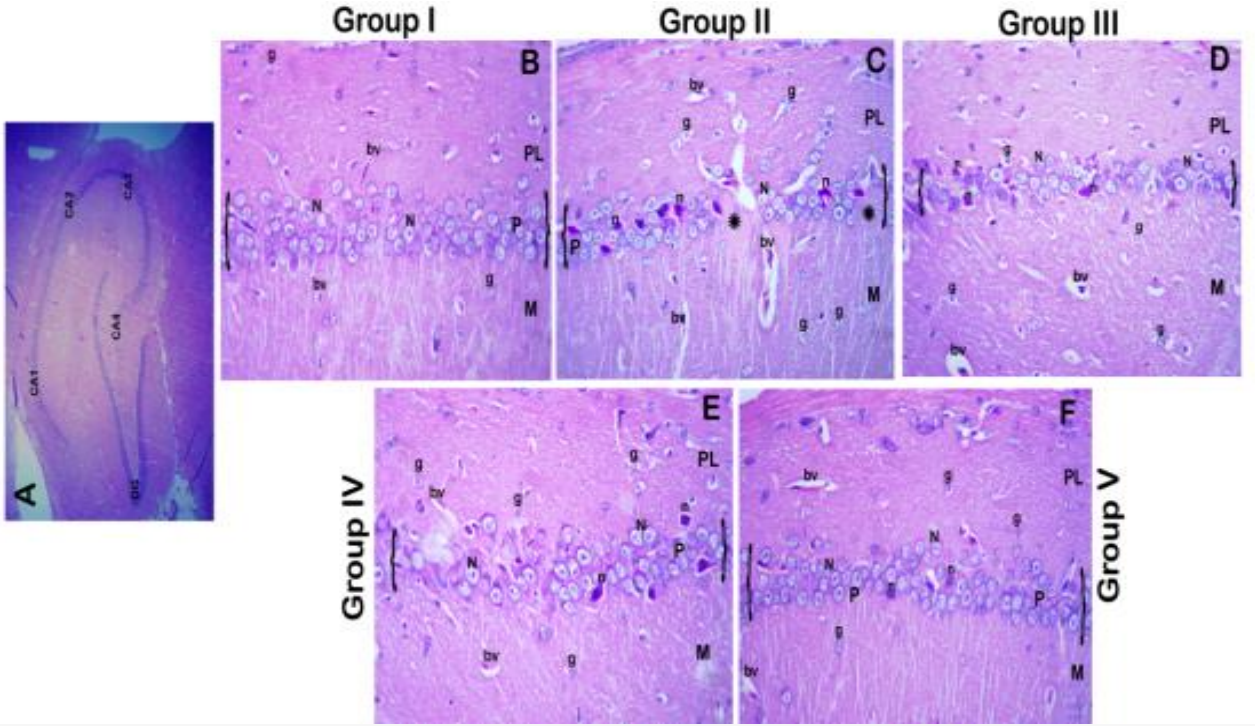

Figure (4): Microphotographs ofCA1 region of hippocampus; from the different studied groups group ,I B ,group II ; C ,group III ;D ,group IV ;E ,and group V F .Haematoxylin-eosin staining (H & E) x400.

DISCUSSION

Our hypothesis was based on the idea that routine blood measures in AD patients could have diagnostic and prognostic value, given the role inflammation plays in the aetiology of the illness. Several inflammatory biomarkers showed distinct differences between the three groups in the current investigation. Significant decreases in the lymphocyte ratio and count values were seen table (3), along with notable variations when compared to the control group. These findings indicate that several pathogenic mechanisms, such as inflammation and oxidative stress, may impact lymphocyte proliferation. The blood biochemistry analysis's findings demonstrated that the positive control group's AChE-RBC concentrations were noticeably greater than those of the control group. There were no discernible variations in glucose, cholesterol, triglycerides, or HDL values in the control and positive control groups. Acetylcholinesterase (AChE), a component of senile plaque, promotes the formation of amyloid fibrils and the synthesis of the very deadly Aβ-AChE. Studies have shown that the Aβ-AChE complexes have a more significant neurotoxic effect than the Aβ peptide alone, both in *vitro* (using hippocampus neurons) and *in vivo* (using the injection of Aβ peptide into the dorsal hippocampus of rats) [16-17].

The cholinergic system is in charge of learning and memory. The cholinergic neurotransmitter acetylcholine is broken down by the enzyme acetylcholine (AChE). AChE is responsible for transmembrane protein ($Na^+K^+ATPase$) activity, which maintains the integrity of the cholinergic neuron membrane. Cholinergic transmission failure is connected to memory impairment in AD. Strong cholinotoxin aluminum can mimic changes in cholinergic transmission by altering the blood-brain barrier (BBB) [18]. In the present investigation, transmembrane protein activity and AChE were significantly elevated in rats given $AlCl_3$. Consistent with earlier research, betalain treatment of $AlCl_3$ resulted in rat intoxication and decreased transmembrane protein activity and AChE.

The gut-brain axis is modulated by polyphenolic antioxidants, such as dietary plant lignans. This process entails converting the polyphenolic compounds through gut bacterial metabolism into molecules that are physiologically active and neuroprotective, which are referred to as human lignans. These metabolites from gut bacteria have neuroprotective effects on a variety of neurodegenerative illnesses, including Alzheimer's disease (AD). Plant-based meals contain non-flavonoid polyphenolic chemicals called lignans, which are converted into polyphenols by gut bacteria as secondary metabolites [19].

There were a few bi-nucleated hepatocytes (circle) and irregularly dilated sinusoids (s). (B3) Multiple bile ducts (Bd), a clogged dilated portal vein (Pv), and an infiltration of inflammatory cells (IF). Hepatocytes (H) had pale cytoplasm and rounded nuclei. Small, darkly stained nuclei and vacuolated cytoplasm were visible in other hepatocytes (h). Small, darkly stained nuclei and vacuolated cytoplasm are visible in other hepatocytes (h). There are no many bi-nucleated hepatocytes (circles). (A4) Hepatocytes arranged regularly in cords (H) and a congested central vein (Cv).

The current study found that *Origanum majorana* leaves contain high carbohydrate content (41.53%) and dietary fiber (15.54%), followed by moderate protein levels (11.52%). These findings align with previous reports, confirming *O. majorana* as a carbohydrate-rich plant [20].

El Amiri *et al.* [20] reported a similar carbohydrate proportion, reinforcing the current study's data. However, minor variations exist. For instance, **Badran *et al.*** [21] documented slightly lower carbohydrate levels (~38–40%), which could be attributed to environmental factors, plant maturity, and drying methods. The presence of terpenoids, flavonoids, steroids, saponins, and alkaloids in the current study is consistent with **Aitbaba *et al.*** [22] who confirmed *O. majorana* as a rich source of these bioactive compounds. Terpenoids and flavonoids, in particular, are known for their antioxidant and anti-inflammatory properties. However, the current study found higher alkaloid levels compared to some reports, such as **Soliman's study** [23], which detected lower concentrations. Differences in extraction methods, solvents, and geographical origins might explain this discrepancy.

The current study identified catechin, quercetin, ellagic acid, and cinnamic acid using HPLC analysis. These findings align with **Imtara *et al.*** [24] who also detected these compounds in *O. majorana*. Additionally, chlorogenic acid and propyl gallate were noted, which contribute to the plant's antioxidant capacity. However, the detection of vanillin and cinnamic acid in the current study is not widely reported, suggesting potential variations in plant chemotype or differences in analytical methods [24]. The current study demonstrated significant improvements in hematological parameters, including RBC count, hemoglobin concentration, and packed cell volume. These findings are supported by **El-Hak *et al.*** [25] who observed similar hematological benefits following *O. majorana* supplementation in animal models. Additionally, **Abou-Seif *et al.*** [26] confirmed that *O. majorana* modulates hematological indices positively. However, the current study reported an increase in platelet count in certain groups, whereas **Soliman *et al.*** [23] observed no significant change. This discrepancy may be due to differences in dosage, exposure duration, or extraction methodology.

Histopathological analysis in the current study revealed alterations in liver hepatocytes, including inflammatory cell infiltration and bile duct irregularities in certain groups. These findings are consistent with **Abou-Seif *et al.*** [26] who reported dose-dependent hepatic changes in *O. majorana*-treated animals. Some studies suggest that higher doses of *O. majorana* extracts may lead to mild hepatotoxic effects, which

could explain the current study's observations. Additionally, **Rababa'h *et al.* [27]** found that *O. majorana* exerts hepatoprotective effects against oxidative stress, supporting the notion that its impact is dose-dependent.

CONCLUSION

There is no clinically available AD disease-modifying treatment currently. But since the beginning of human history, medicinal plants have been used to treat ailments. In this study, *Origanum majorana* was found to improve brain function and may have a role in treating Alzheimer's disease (AD) due to its high content of natural compounds with anti-inflammatory, antioxidant, and anticholinesterase activities such as flavonoids, polyphenols, triterpenes, sterols, and alkaloids. In this study, a few inflammatory biomarkers revealed notable variations among the three cohorts. The lymphocyte ratio and count values were significantly lower in the experimental group compared to the control group, indicating the possibility of many pathogenic processes influencing lymphocytes.

Conflict of interest: None.
Financial disclosures: None.

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