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Effects of Ellagic Acid Supplementation on Antioxidant Status and Symptom Improvement in Patients with Major Depressive Disorder: A Double-blind Randomized Clinical Trial

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Abstract

Background: Depression is one of the most common mood disorders and a major public health concern. Ellagic acid (EA), a type of polyphenol, acts as a strong hydrogen bond network as an electron receptor, enabling it to participate in various reactions.
Objectives: Major depression is a critical medical condition that has emerged as a public health issue due to its high incidence, mortality, and suicide rates. One significant factor in the pathogenesis of depression is oxidative stress. This study aimed to evaluate the effects of ellagic acid supplementation on antioxidant status and symptom improvement in patients with major depressive disorder, considering the antioxidant capabilities of ellagic acid.
Methods: A total of 40 patients diagnosed with major depressive disorder based on DSM-V criteria were assessed using the Beck Depression and Hamilton Depression Questionnaires. The dietary and caloric intake of the patients were monitored. Additionally, height and weight were measured, and patients with similar age, gender, and weight were matched. The individuals were randomly assigned to either the intervention group or the placebo group using a randomization table. The intervention group received a daily dose of 200 mg of ellagic acid in capsule form. The placebo group received a daily dose of one capsule containing 200 mg of wheat flour, identical in appearance to the intervention substance. The study period lasted for 8 weeks. Venous blood samples were collected before and after the study from all 40 individuals, and after serum separation, oxidative stress markers (malondialdehyde and total antioxidant capacity). were measured using a specific kit and ELISA method.
Results: The study results showed a significant reduction in depression scores in the ellagic acid group during the study (P: 0.001)., with these alterations being significant when compared to the placebo group. In the ellagic acid group, a significant increase in total antioxidant capacity (P: 0.027). and a significant decrease in malondialdehyde levels (P: 0.014). were observed at the end of the study, and these changes were significant compared to the placebo group. In contrast, significant changes in total antioxidant capacity and malondialdehyde levels were not observed in the placebo group.
Conclusions: The current study indicates that ellagic acid intervention may have a favorable effect on depression in patients with major depressive disorder. This is achieved by reducing BDI scores and serum levels of MDA, as well as increasing serum levels of TAC in these patients compared to the placebo group. However, further investigation is necessary to explore the mechanisms underlying the different alterations of ellagic acid in depression.

Keywords: Major Depressive Disorder, Ellagic Acid, Double-blind Randomized Clinical Trial, Total Antioxidant Capacity, MDA

1. Background

Depression is one of the most common mood disorders and a major public health concern (1). It carries the highest burden of non-communicable diseases, with 12% of an individual's life being affected by disability due to this condition, and it was considered the third leading cause of disability worldwide in 2020 (2, 3). It affects 25% of women and 12% of men and has a significant prevalence in Iran (4, 5). Major depressive disorder occurs without a history of manic, mixed, or hypomanic episodes and must last for at least two

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weeks. To diagnose major depressive disorder, the patient must have at least four symptoms from a list, including changes in appetite and weight, changes in sleep and activity, loss of energy, feelings of guilt, difficulty thinking and making decisions, and recurrent thoughts of death or suicide (6).

Depression is associated with adverse effects such as disability and anxiety, decreased fertility, decreased cognitive ability, increased mortality, and overall reduced quality of life (7-9). The causes of mood disorders are not fully understood, but they can be classified into biological, genetic, social, and psychological factors (6). Several studies have shown a close relationship between depression and neurotransmitter activity in the central nervous system, such as serotonin, norepinephrine, and dopamine (10). According to this hypothesis, a decrease in the levels of monoamines (serotonin, norepinephrine, and dopamine). in the brain can lead to depression (11). Depression is accompanied by a decrease in the levels of neurotransmitters such as serotonin, norepinephrine, and dopamine in the cortical regions of the brain and the limbic system. Most antidepressant medications available on the market have a significant effect on the amino acid system. The neurotransmitters norepinephrine and serotonin have the greatest involvement in the pathophysiology of mood disorders. Depression is known as a major emotional stressor. After stress occurs in the body, catecholamines are released, leading to an increase in free radicals. The monoamine hypothesis of depression, proposed by Schildkraut in 1965, suggests that depression is caused by a deficiency in the function of monoamine transporters in certain areas of the brain (12-14). Oxidative stress is an imbalance between free radicals and antioxidant defenses, leading to the oxidation of proteins, lipids, and nucleic acids. High oxidative stress or low antioxidant status in biology indicates poor health outcomes and a wide range of diseases (15, 16).

Ellagic acid (EA). is one of the types of polyphenols discovered by Braconnot in 1831 (17). It acts as a strong hydrogen bond network as an electron receptor, which enables ellagic acid to participate in some reactions. This polyphenol is naturally present in many fruits and vegetables, including strawberries, red and black currants, and nuts such as walnuts, pistachios, almonds, oak nuts, pomegranates, and grapes, especially in their seeds and skins (18). Ellagic acid has a wide range of pharmacological activities such as antioxidant, anticancer, anti-allergic, anti-malaria, and anti-inflammatory effects. Additionally, ellagic acid has neuroprotective activity against oxidative damage (19-22). In summary, the anti-inflammatory mechanism of ellagic acid is associated with the reduction of malondialdehyde (MDA). levels, inducible nitric oxide synthase (iNOS)., and 2COX- by suppressing pro-inflammatory cytokines (TNFα and IL1β). It has been shown that ellagic acid regulates NOS and COX to suppress the release of NO and prostaglandin E2, which are inflammatory mediators (23, 24).

2. Objectives

The aim of this study is to investigate the effects of ellagic acid supplementation on antioxidant status and the improvement of symptoms in individuals with major depressive disorder.

3. Methods

3.1. Study Design

The present study was a single-center, double-blind, randomized, placebo-controlled clinical trial. This study was approved by the Research Ethics Committee of Mazandaran University of Medical Sciences, Sari, Iran (Approval ID: IR.MAZUMS.REC.1399.697; Approval date: 2020/09/16). The study protocol was prospectively registered in the Iranian Registry of Clinical Trials (IRCT). at irct.ir with the Clinical Trial Code IRCT20141025019669N17. The study was conducted following the principles of the Helsinki Declaration.

All patients who entered the trial were randomized, in a double-blind design, to receive either a placebo (n = 20). or a capsule containing 200 mg of ellagic acid daily for 8 weeks. The placebo capsules contained 200 mg of wheat flour, identical to the ellagic acid capsules in dimensions, weight, smell, color, and form. The placebo was produced by the School of Pharmacy at Mazandaran University of Medical Sciences (Sari, Iran)., and the supplement was purchased from Exir Gostar Espadana. It should be noted that Kazemi et al. determined the optimal dosage for ellagic acid supplementation, and this dosage was chosen because oral ellagic acid supplementation has exhibited antioxidant properties, which was the main objective of conducting this study (25).

3.2. Patients

The study included patients with major depression who visited a psychiatric clinic. Ethical approval was obtained from the university's ethics committee, and informed consent was secured from interested individuals. Patients with major depression were identified using structured clinical interviews and diagnostic criteria from DSM-V, as well as depression questionnaires, with a depression score above 20. Clinical diagnosis and testing were performed by a clinical consultant (psychiatrist).

3.3. Inclusion Criteria

Patients meeting the following criteria were included in the study: Consent to participate, diagnosis of major depression according to DSM-V diagnostic criteria and a depression score above 20, age over 18 years, fluoxetine consumption (doses of 20 to 60 milligrams)., and no use of any supplements for at least 2 weeks.

3.4. Exclusion Criteria

The exclusion criteria for this study were: Pregnancy and lactation, patients with kidney and liver disorders, any acute illness that may affect the study (cardiovascular, pulmonary, renal, cancer patients)., lack of interest in participating in the study, history of any allergies, smoking, alcohol consumption, travel, occurrence of any side effects due to the intervention, and lack of interest in participating or continuing to cooperate.

3.5. Randomization and Blinding

To ensure allocation concealment, the Sequentially Numbered, Opaque, Sealed Envelopes (SNOSE). method was used in this study. This common method involves generating a random sequence using specialized software and preparing aluminum-wrapped envelopes to prevent the contents from being visible. The number of envelopes was based on the sample size of the study, and each envelope contained a registration card with a recorded random sequence. To maintain the integrity of the random sequence, the envelopes were numbered on the outer surface in the same order. Finally, the envelopes were sealed and placed in a box in order. During participant enrollment, an envelope was opened based on the order of entry of eligible participants, and the assigned group was revealed.

3.6. Laboratory Methods

Forty venous blood samples were collected at the beginning and the end of the 8-week intervention. After a 15-minute centrifugation at 2000 g, the serum samples were frozen at -80°C for future laboratory assessment. The serum levels of malondialdehyde (MDA). and total antioxidant capacity (TAC). were evaluated using commercial kits (Human ELISA; Kushan Zist Azma Parseh). according to the manufacturer's protocol. MDA in the samples reacts with thiobarbituric acid (TBA). in an acidic environment, producing a pink-colored MDA-TBA adduct, which can be easily measured photometrically (530 - 540 nm). or fluorometrically (Ex/Em = 532/553 nm). Additionally, the TAC assay kit measures TAC in biological samples by employing a peroxidase chromogenic substrate (ABTS). that generates a water-soluble (blue-green). chromogen upon oxidation by ferryl myoglobin radicals. The rate of generation of the colored chromogen is repressed by the presence of antioxidants and can be quantified photometrically.

3.7. Instruments

A validated Beck questionnaire was used to determine the depression state of each MDD participant before and after the supplement intervention through interviews conducted by a psychiatrist to determine the score and severity of depression. The Beck Depression Inventory (BDI). consists of a 21-question self-rating depression scale. Each question has four answers, ranging from low to high, representing the degree of depression. After reading the questions in order, the participants select the options that best describe their moods during the last two weeks. The overall depression score determined by the 21 options indicates: Healthy Status (1 - 18)., Mild Depression (18 - 28)., Moderate Depression (29 - 35)., and Severe Depression (36 - 63) (26, 27).

In addition, the severity of the symptoms was assessed using the Hamilton Depression Rating Scale (HAM-D). Patients who had already received a diagnosis of depression were asked to measure the severity of their symptoms using the HAM-D. The HAM-D comprises seventeen questions that allow the rater to evaluate each symptom's intensity on either a five-level (0 to 4).

or a three-level (0 to 2). scale. The patient's state is represented by the score: 0 to 6 shows no indications or symptoms of depression; 7 to 17 indicates mild depression; 18 to 24 indicates moderate to severe depression. A score of 24 or above indicates serious depression (28, 29).

3.8. Nutritional Evaluation

The food recall questionnaire included a three-day dietary recall, which also covered one day off, and was completed at the beginning, fourth week, and end of the study. The food recall data were analyzed using Nut 4 software.

3.9. Patient Follow-up

Patient follow-up was conducted via telephone contact every 7 days to monitor ellagic acid capsule consumption and prevent any loss of capsules. Compliance was evaluated by counting the remaining capsules at the end of the eighth week. Patients who had not consumed at least 90% of their supplement were excluded from the study.

3.10. Sample Size

To calculate the sample size, the depression score variable from the study by Khajehnasiri et al (30). was used, which investigated the effects of vitamin C antioxidant administration. In this study, the mean and standard deviation of the depression score before vitamin C supplementation were 14.133 ± 4.37, and after supplementation, they were 7.08 ± 3.233. Based on these values, a sample size of 12 participants per group, with 95% probability and 99% confidence level, was determined to be sufficient to reject the null hypothesis of equal effect before and after the study.

$N = [(Z1-\alpha/2 + Z1-\beta)\ 2\ (SD12+SD22)]\ /\Delta 2$; ($Z1-\alpha/2 = 2.58$, $Z1-\beta = 1.64$; $N = 12$).

Considering the potential for drop-outs, the sample size was increased to 22 participants per group. Thus, a total of 44 pre-diabetic patients were included in this study.

3.11. Statistical Analysis

Data were presented as mean (± standard deviation). for quantitative variables and as frequency (percentage) for qualitative variables. The normality of data distribution was evaluated using the Kolmogorov-Smirnov test. For quantitative variables with a normal distribution, the paired t-test was used to compare means within each group, and the Student *t*-test was used to compare means between the two groups. For non-normally distributed data, the Wilcoxon test was used for within-group comparisons, and the Mann-Whitney test was used for between-group comparisons.

Efforts were made to match the two groups receiving the supplement and placebo as closely as possible. Any differences between the groups were controlled for through covariance analysis to eliminate the effects of confounding factors. A P value of less than 0.05 was considered statistically significant. Statistical analysis was performed using SPSS version 20 software without knowledge of the treatment group.

4. Results

4.1. Demographic Characteristics

In this trial, 44 people were included, with two participants from each group (intervention and placebo). dropping out due to personal reasons, resulting in a 90 percent cooperation rate. A total of 40 patients (20 in each group). completed the study. The mean age was 54.18 years in the intervention group and 53.29 years in the placebo group. The results showed no significant differences in mean age, height, and BMI between the two groups (Table 1).

4.2. Dietary Comparison

The results showed no significant difference in dietary intake between the two groups at the beginning of the study. Additionally, there were no significant changes in these parameters during the study in either group. At the end of the study, after adjusting for baseline values, there remained no significant difference between the two groups (Table 2).

4.3. Depression Scores

The study showed a significant decrease in depression scores in the ellagic acid group (P = 0.001). However, the reductions in the placebo group were not significant. While depression scores did not differ significantly between the two groups at the beginning of the study, after adjusting for baseline values, they were significantly lower in the ellagic acid group compared to the placebo group at the end of the study.

Table 1. Demographic Characteristics

Variables	Placebo (n = 20)	Ellagic Acid (n = 20)	P1
Age	53.29 ± 6.19	54.18 ± 5.85	0.601
Height (cm)	165.42 ± 10.29	164.55 ± 11.08	0.792
Weight (kg)			
Before	70.08 ± 8.11	71.28 ± 9.17	0.408
After	69.58 ± 9.04	70.46 ± 9.17	0.328
P2	0.382	0.4	
BMI (Kg/m²)			
Before	25.61 ± 0.77	26.32 ± 0.75	0.603
After	25.42 ± 0.86	26.02 ± 0.56	0.586
P2	0.509	0.521	
Physical activity			
Before	32.17 ± 3.09	31.05 ± 3.33	0.101
After	33.54 ± 3.29	31.77 ± 4.58	0.091
P2	0.112	0.254	

[a] Variables are expressed as Mean ± SD.

In the ellagic acid group, the reduction in Beck's Depression Inventory was about 43%, and in Hamilton's Depression Scale, it was about 48% (Table 3).

4.4. Oxidant and Antioxidant Evaluation

Ten venous blood samples were taken at the beginning and end of the study. After serum separation, oxidative stress markers (malondialdehyde and total antioxidant capacity). were measured in the plasma of participants using a specific kit and ELISA method. In the ellagic acid group, a significant increase in TAC (total antioxidant capacity). level was observed at the end of the study ($P < 0.05$), while no significant change was observed in the TAC level in the placebo group. Additionally, a significant decrease in MDA (Lipid Peroxidation Index). level was observed in the ellagic acid group at the end of the study ($P < 0.05$)., while no significant change was observed in the MDA level in the placebo group. In the ellagic acid group, TAC increased by about 30%, and MDA decreased by about 49% (Table 4).

5. Discussion

The results of the current study showed that ellagic acid supplementation significantly reduced depression scores in patients with major depression. There was no significant difference in baseline characteristics between the two groups. Moreover, there was no significant difference in dietary intake between the two groups at the beginning of the study. Additionally, there was no significant change in these parameters during the study in either group, and at the end of the study, after adjusting for baseline values, there was no significant difference between the two groups. The study demonstrated that ellagic acid supplementation significantly reduced blood MDA levels and oxidative stress in patients with major depression compared to the placebo group. Furthermore, ellagic acid supplementation significantly increased blood TAC and total antioxidant levels in patients with depression compared to the placebo group.

Highly reactive substances known as reactive oxygen species (ROS). are produced by a cell's regular metabolic activity. These hazardous oxygen species can damage lipids, proteins, and DNA, among other biomolecules. The carcinogenic and toxic chemical malondialdehyde (MDA). is generated when arachidonic acid is oxidized. Numerous disorders, including major depressive disorder (MDD)., can disrupt the equilibrium between antioxidant defense and free radicals (31). Several clinical trials have linked depressive disorders to oxidative stress in the brain and blood (32, 33). Additionally, it has been shown that patients diagnosed with major depression have decreased antioxidant enzyme activities, which are mitigated by antidepressant treatment (34). Ellagic acid has been shown to exhibit antioxidant activity (35)., which may also account for its antidepressant-like activity in experimental reports (36). Its free radical scavenging action, which has been compared to that of vital

Table 2. Dietary Comparison

Variables	Placebo (n = 20)	Ellagic Acid (n = 20)	P1
Energy(kcal)			
Baseline	2308.45 ± 265.09	2381.08 ± 303.25	0.65
End	2300 ± 250.058	2335.84 ± 289.11	0.78
P2	0.809	0.711	
Protein(gr)			
Baseline	74.07 ± 14.2	75.22 ± 19.04	0.401
End	73.86 ± 22.11	75.09 ± 27.31	0.238
P2	0.356	0.569	
Carbohydrate(gr)			
Baseline	295.47 ± 58.04	314.18 ± 39.05	0.44
End	294.29 ± 39.44	311.28 ± 45.44	0.508
P2	0.699	0.59	
Fat (gr)			
Baseline	92.5 ± 16.22	92.3 ± 15.07	0.419
End	90.18 ± 24.18	91.17 ± 15.07	0.488
P2	0.309	0.41	
Saturated fatty acid(gr)			
Baseline	30.05 ± 7.19	29.85 ± 6.39	0.219
End	29.24 ± 5.34	29.11 ± 6.44	0.394
P2	0.207	0.35	
Monounsaturated fatty acids (gr)			
Baseline	32.13 ± 5.77	30.14 ± 6.22	0.313
End	30.07 ± 4.28	29.17 ± 5.66	0.396
P2	0.274	0.402	
Polyunsaturated fatty acids (gr)			
Baseline	28.46 ± 5.93	29.27 ± 7.11	0.257
End	27.65 ± 6.08	29.07 ± 7.03	0.143
P2	0.199	0.29	
Fiber (gr)			
Baseline	13.29 ± 2.53	14.69 ± 2.58	0.28
End	13.01 ± 3.19	13.88 ± 3.12	0.294
P2	0.49	0.215	
Vitamin C (mg)			
Baseline	72.25 ± 19.23	73.18 ± 12.44	0.506
End	71.5 ± 20.33	73 ± 24.03	0.397
P2	0.466	0.701	
Vitamin E (IU)			
Baseline	11.59 ± 2.33	12.77 ± 3.04	0.096
End	10.46 ± 2.28	11.99 ± 2.22	0.871
P2	0.183	0.104	
Selenium			
Baseline	115.28 ± 37.14	118.27 ± 44.1	0.425
End	114.95 ± 37.6	118.33 ± 61.07	0.303
P2	0.661	0.746	

[a] Variables are expressed as Mean ± SD.

vitamins ascorbic acid and α-tocopherol, is attributed to its inherent antioxidant qualities. Ellagic acid can scavenge a broad range of ROS and reactive nitrogen species (RNS). due to its two lactone and four hydroxyl functional groups (37).

Table 3. Depression Scores

Variables	Placebo (n = 20)	Ellagic Acid (n = 20)	P1
Depression-Beck			
Baseline	17.95 ± 4.21	18.33 ± 4.19	0.298
End	17.65 ± 4.11	10.4 ± 3.5	0.001
P2	0.312	0.001	
Depression- Hamilton			
Baseline	17.5 ± 4.47	17.89 ± 3.33	0.32
End	17.11 ± 3.93	9.16 ± 3.24	0.001
P2	0.284	0.001	

[a] Variables are expressed as Mean ± SD.

Previous research performed by Taene et al. demonstrated a statistically significant reduction in the serum TAC level and a significant elevation in the serum MDA level in MDD patients compared to healthy individuals (31), which is consistent with our research. However, their results showed a positive correlation between the severity of depression and the level of MDA serum. Furthermore, they found a meaningful negative association between the severity of the disease and serum TAC level in MDD patients versus healthy volunteers. The authors also indicated that the NLRP3 inflammasome was significantly upregulated in MDD patients compared to healthy volunteers. Previous reports focused on the crucial role of psychological stress in contemporary life as a danger signal that activates the inflammasome and releases cytokines, sending inflammatory signals to the brain (31).

The results of this study are consistent with those of the study by Ghadimi et al., which investigated the effect of ellagic acid on blood glucose levels, insulin resistance, lipid profile, oxidative stress, antioxidant levels, and inflammatory factors in patients with type 2 diabetes. The authors indicated that the blood MDA level in the ellagic acid group significantly decreased, and the blood TAC level meaningfully increased at the end of the study (38). Few investigations have been conducted to demonstrate the antidepressant activity of ellagic acid. One study revealed that the administration of ellagic acid (180 mg for 12 weeks). to multiple sclerosis patients with mild to moderate depressive symptoms produced a favorable effect on depression in these patients. This was achieved by lowering nitric oxide (an oxidative marker)., cortisol (a stress hormone)., and indoleamine 2, 3-dioxygenase (a possible indicator of inflammation and oxidative stress). gene expression levels, in addition to Beck Depression Inventory-II scores. Moreover, the authors demonstrated a substantial increase in serum levels of serotonin and BDNF. Nevertheless, at the end of the investigation, there were no substantial variations in serum Nrf2 (an important component of the antioxidant defense). levels between the treatment and control groups (39). These findings suggest a complex and crucial role for ellagic acid in the pathophysiology of depression. Therefore, further investigation is necessary to explore the mechanisms underlying the different effects of ellagic acid in depression.

Cervantes-Anaya et al., Ferreres et al., and Dhingra et al. have found that part of the antidepressant activity of ellagic acid may be related to its antioxidant and free radical-scavenging activities in vitro and in vivo models of depression (40-42). Cervantes-Anaya et al. assessed the impact of the main compounds found in the aqueous extract of pomegranate (such as ellagic acid). on the redox environment in ovariectomized rats, and like our results, they reported that ellagic acid improved the antidepressant-like profile and antioxidant activity by mitigating oxidative damage (40). Ferreres et al. investigated the antidepressant, anti-cholinesterase, and antioxidant properties of ellagic acid and a medicinal plant extract in vitro. The authors revealed that ellagic acid has a greater ability than vitamin C to scavenge free radicals (superoxide anion and nitric oxide radicals). In addition, ellagic acid indicated strong antidepressant activity, which could perhaps be associated with its antioxidant properties. Their findings indicated that ellagic acid and its analogues are less potent inhibitors of cholinesterase enzymes. However, as a selective inhibitor of monoamine oxidase A (MAO-A)., ellagic acid has been shown to have strong antidepressant potential. MAO-A acts by increasing synaptic monoamine concentrations and preventing the overproduction of hydrogen peroxide, an end

Table 4. Oxidant and Antioxidant Evaluation

Variables	Placebo (n = 20)	Ellagic Acid (n = 20)	P1
TAC (mg/dL)			
Baseline	1.81 ± 0.17	1.79 ± 0.1	0.255
End	1.9 ± 0.11	2.56 ± 0.19	0.027
P2	0.129	0.014	
MDA			
Baseline	1.79 ± 0.33	1.83 ± 0.2	0.151
End	1.75 ± 0.28	0.92 ± 0.14	0.014
P2	0.11	0.02	

[a] Variables are expressed as Mean ± SD.

product of the deamination reaction that MAO-A catalyzes (41). This project may prove that ellagic acid exerts its antidepressant effects through multiple pathways. Additionally, Girish et al. demonstrated that ellagic acid plays an important role in its antidepressant-like activity in mice through its impact on the monoaminergic system (serotonergic and noradrenergic systems) (43). In the study by Dhingra and Chhillar, which investigated the antidepressant effects of ellagic acid on stressed and unstressed mice, the results showed that ellagic acid had significant antidepressant effects on unstressed mice, possibly by stimulating and activating the adrenergic and serotonergic systems. However, in stressed mice, they suggested that ellagic acid may have antidepressant-like effects by inhibiting inducible NOS (42).

In the study by Lorigooini et al., the antidepressant effects of ellagic acid were studied in male mice through the NMDA-NO pathway. According to the results, ellagic acid exerted antidepressant effects in male mice by affecting at least part of the NMDA-NO pathway. This study investigated the antidepressant effects of ellagic acid through a different neural pathway, which was inconsistent with our report (44). Mise Yonar et al. evaluated the effects of ellagic acid on immunological, hematological, and antioxidant parameters in rainbow trout. The results showed that ellagic acid had a significant effect on the antioxidant activity of the fish and increased the total antioxidant level, which is consistent with the findings of the present study (45).

In the review study by Jalali et al., the effects of traditional Iranian herbal medicines on improving depressive symptoms were studied. According to the results, ellagic acid present in tannins had a significant effect on improving depressive symptoms and could be effective for future pharmacogenetic studies in managing depression. The results of this study were consistent with the present study in terms of the antidepressant effects of ellagic acid supplementation (46). In the study by Hassonizadeh Falahieh et al., the effects of ellagic acid supplementation on anxiety, depression, motor behaviors, blood-brain barrier permeability, brain edema, and inflammation in male rats were investigated. Sixty rats were randomly divided into six groups of ten. Cerebral ischemia/reperfusion was induced by occluding bilateral common carotid arteries for 20 minutes, and then reperfusion was initiated. Behaviors were tested one week after treatment, and brain tissue cytokines were measured using specific ELISA kits. The results showed that ellagic acid supplementation, possibly through its anti-inflammatory effects, could be effective in improving anxiety, depression, motor behaviors, edema, and brain inflammation, and could be used as an effective therapeutic agent against cerebral ischemia/reperfusion. The results of this study were consistent with the findings of the present study (47).

5.1. Safety and Adverse Events

No side effects were reported in the study. Additionally, this study did not include any co-interventions.

5.2. Limitation Section

Despite the encouraging outcomes of our study, it is crucial to consider its limitations. One limitation is the absence of evaluation of other stress markers, including ROS, hydroxyl radical (·OH), superoxide anion (O2·−)., NO, and peroxynitrite (ONOO−), as well as the activity of glutathione peroxidase (GPx). and superoxide

dismutase (SOD), and serum nuclear factor erythroid-2-related factor 2 (Nrf2). Another limitation is the lack of a correlation analysis to indicate the relationship between antioxidant activity and the antidepressant-like activity of ellagic acid.

Furthermore, because major depressive disorder (MDD). Is complex and heterogeneous, multiple etiologies, including the biogenic amine hypothesis, receptor hypothesis, cytokine theory, neurotrophic factor hypothesis, and others, may contribute to the development of depression (48). Few investigations have been performed to display the antidepressant-like activity of ellagic acid in animal models. Their results showed that ellagic acid treatment could affect selective serotonin reuptake, monoaminergic neurotransmitter receptors, BDNF, and the nitric oxide (NO) system (39, 42, 43). Nevertheless, no clinical research has been carried out to support ellagic acid's potential antidepressant effects in cases of major depression via the above-mentioned mechanisms. This is a complementary therapy in addition to a lifestyle adjustment. Thus, long-term cohort studies could provide more information about the antidepressant-like activity of ellagic acid and its underlying mechanisms in MDD patients.

5.3. Conclusions

Our double-blind randomized clinical trial showed that, in patients with a history of MDD, ellagic acid supplements significantly reduced oxidative stress and elevated serum levels of total antioxidant capacity (TAC). However, significant changes in TAC and MDA levels were not observed in the placebo group. Additionally, the results showed no significant difference in dietary intake between the two groups (placebo and intervention) at the beginning and end of the study. Given that major depressive disorder is complex and heterogeneous, long-term cohort studies incorporating multi-component lifestyle interventions should be undertaken to provide more information about the antidepressant-like activity of ellagic acid and its underlying mechanisms (monoamine neurotransmitter levels, inflammation, the BDNF/tyrosine kinase B (TrkB) signaling pathway, etc.) in MDD patients.

Footnotes

Authors' Contribution: Conception and design: S.H.H, M.Z, H.K.H, and G.H; drafting the article: H.K.H, A.B, S.A.K, and S.K.S; critically revising the article: G.H; approved the final version of the manuscript: All authors.

Clinical Trial Registration Code: IRCT20141025019669N17 .

Conflict of Interests Statement: None of the researchers involved in this study have any financial or non-financial conflicts of interest, no personal financial interest is reported. Dr. Houshmand and Dr. Hosseini are among the reviewers of this study, while Dr. Zarghami is a member of the editorial board.

Data Availability: The dataset presented in the study is available on request from the corresponding author during submission or after its publication. The data are not publicly available due to the very high volume of results, if we wanted to publish all the results in article format, a very large volume would be occupied.

Ethical Approval: IR.MAZUMS.REC.1399.697 .

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