JHE JOURNAL OF HOME ECONOMICS, MENOUFIA UNIVERSITY
Website: https://mkas.journals.ekb.eg
Print ISSN 2735-5934 Online ISSN 2735-590X

Nutrition and Food Sciences
Article Type: Original article

Received: 5 Feb 2025
Accepted: 16 Mar 2025
Published: 1 Apr 2025

The Effect of Dietary Intervention with Pomegranate and Aloe Vera in Alleviating the Symptoms of Alcohol-Induced Stomach Ulcers in Experimental Rats

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ABSTRACT:

Peptic ulcer disease is defined by open lesions in the stomach and duodenum, frequently associated with oxidative stress and inflammation. This study evaluated the therapeutic potential of a probiotic functional formula enriched with pomegranate peel extract and aloe vera gel. Thirty-five adult male rats were allocated into a negative control group (n=7 rat) and ulcer-induced groups (n=28 rat), which received a single oral dose of ethyl alcohol at a concentration of 10 ml/kg. The ulcer-induced groups were further categorized into four equal subgroups, each receiving distinct treatments: 200 mg/kg of aloe vera (AVG), 50 mg/kg of pomegranate (PMG), and a combined treatment of aloe vera and pomegranate (APG). The results indicated that the dietary interventions significantly improved hematological and antioxidant parameters. Notably, the APG group demonstrated the highest hemoglobin level (12.25±0.70 g/dl) and red blood cell (RBC) count (7.03±0.59), representing a substantial enhancement compared to the ulcer-induced control group. Additionally, antioxidant activity was markedly enhanced, with the APG group exhibiting the highest superoxide dismutase (SOD) level (25.03±0.55 u/ml) and lower malondialdehyde (MDA) levels (1.54±0.53 nmol/ml). Furthermore, the ulcer inhibition index was highest in the APG group (359.71±44.14), which also recorded the greatest protection rate (60.17±5.24%) and the lowest ulceration percentage (35.87±5.89%). These findings underscore the synergistic protective effects of aloe vera and pomegranate in mitigating gastric damage, enhancing antioxidant defenses, and promoting ulcer healing. The combined treatment exhibited significant therapeutic benefits, suggesting its potential as a natural approach for alleviating peptic ulcers and improving overall gastrointestinal health.

Keywords: Gastric, Prebiotic, Antioxidant, Inflammation, Protective index, Ulcer score

Cite as: Gharib et al., 2025, The effect of dietary intervention with pomegranate and aloe vera in alleviating the symptoms of alcohol-induced stomach ulcers in experimental rats. JHE, 35 (2), 57-84. DOI: 10.21608/mkas.2025.365849.1373

1. INTRODUCTION

Peptic ulcer disease (PUD) is a type of gastrointestinal ulcer that develops on the stomach lining or the upper portion of the small intestine and is linked to damage in the gastric mucosa and submucosa of the stomach and duodenum. It happens when there is an imbalance between defensive elements like gastric mucus and antioxidant defenses and

aggressive elements like stomach acid and free radicals. Inflammation and oxidative stress are the two main processes behind stomach ulcers. [1].

To eradicate peptic ulcers, ongoing development into more effective antiulcerogenic medications is crucial, depending on availability and cost. According to research, a variety of fruits and vegetables have anti-inflammatory, antioxidant, anti-secretory, antibacterial, anticholinergic, and cellular defense mechanisms that can prevent peptic ulcers. Furthermore, the phytochemicals included in fruits and vegetables are essential for both illness prevention and therapy. [2].

As a result, research was conducted on aloe vera, another readily available tropical plant that may help those who suffer from ulcers. The Arabic word "Alloeh" means "shining bitter substance," and the Latin word "vera" means "true." These are the origins of aloe vera. Aloe was known to the Egyptians as "the plant of immortality." For millennia, aloe plants have been utilized as medicine. One of the most popular medicinal plants in human history is Aloe barbadensis (1,8-Dihydroxy-3-hydroxymethyl-10-(6-hydroxymethyl3,4,5-trihydroxy-2-pyranyl anthrone), often known as aloe vera. [3]

Aloe Vera is a small, stemless plant that grows between 60 and 100 cm in height. Varieties with white spines have green, thick, meaty leaves [4]. The blooms, which are 90 cm tall pendulums with yellow tubular corollas, are mostly produced in the summer. The phytochemical makeup consists of lectins, anthrones, and anthraquinone c-glycosides. It has been discovered that giving aloe vera within seven days caused the stomach ulcer to heal. There is a reduction in the stomach acid production particularly in the high dose group as compared with the omeprazole group, in line with the prior study in acid output reduction [5]. Prostaglandins, fatty and amino acids, are thought to be the mechanism of action of acid reduction [6]. The small rise in stomach acid production in the Aloe vera treatment group may be explained by the tendency of amino acids in protein to raise hydrochloric acid levels to convert inactive pepsinogen to pepsin. Gibberellin, auxins, and fatty acids have also been discovered to be present in aloe vera [7]. Auxin and gibberellin promote wound healing, while fatty acids raise prostaglandin levels, which control inflammation. In contrast to its high dosage, aloe vera reduces acid output with increased use, as seen on day 28. It indicates that, as was shown in the earlier work using a tropical plant, a periodic rise improves prostaglandin production for mucus protective manufacture [8]. From day 7 to day 28 of the trial, it was demonstrated that prostaglandin levels rose, which aided in the gastric ulcer's healing. This suggests that during the healing process, strong herbal treatments enhance the prostaglandin rise pathway through the production of fatty acids.

In ancient folk medicine, pomegranates (Punica granatum) were used to prevent or treat a variety of stomach and intestinal disorders. Numerous active substances, including flavonoids (catechins and quercetin), phenolic compounds (gallagic acid), and anthocyanidin (cyanidin), were present in all regions of pomegranates [9], They have been shown to have a variety of biological benefits, including anti-inflammatory, antioxidant, antibacterial, and antiulcer properties [10]. Pomegranate peel (PP) is the inedible film layer inside the pomegranate and its outermost hard structure. It is a by-product of pomegranate products manufacturing. It makes up roughly 30% to 40% of the entire pomegranate and is typically thrown away as waste [11]. Because of its abundance of polyphenols, pomegranate peel extract (PPE) has garnered a lot of interest [12]. Talaat et al. [13] and Chauhan et al. [14] revealed that pomegranate peel aqueous or ethanol extracts had a gastro-protective effect against either Ethier or aspirin-induced gastric ulcer model in rats, indicating that the tannin level

in PP can range from 10.4% to 21.3%. Ifora et al. [15] found that in rats with ethanol-induced stomach ulcers, pomegranate rind extracts reduced inflammation by suppressing the expression of the cyclooxygenase-2 protein. Probiotics are live bacteria that help the host's health when taken in sufficient quantities. One of the most well-known sources of probiotics is yogurt, which contains Bifidobacterium and Lactobacillus strains. By encouraging the growth of good bacteria and suppressing bad bacteria, these probiotics aid in preserving a balanced gut microbiota[16].

It is well known that probiotics lessen intestinal inflammation. One of the main causes of ulcer development and delayed healing is inflammation. Probiotics have the potential to improve ulcer healing rates by reducing stomach lining inflammation through immune response modulation. According to research, probiotics may help ulcers heal by boosting growth factor production and enhancing blood flow to the afflicted areas. This can hasten the healing process for ulcer sufferers [17].

The goal of nutritional therapy is to encourage recovery through a complicated series of steps that start with the original trauma and end with the repair of the injured tissue. Its anti-oxidant, anti-inflammatory, mucus-secreting, cytoprotective, or healing properties may help explain its anti-ulcer properties [3]. Therefore, the purpose of this investigation was to determine the anti-ulcer effects of functional probiotic formulas containing pomegranate and aloe Vera on peptic ulcer induced albino rats.

2. Materials and Methods

2.1 Materials

The fresh pomegranate and aleo vera were gathered from Shiben El-Kom Supermarket in the Menoufia Governorate of Egypt.

Adult male white albino rats were purchased from the Medical Analysis Department of the Research Institute of Ophthalmology in Giza, Egypt.

Cellulose, casein, and a blend of vitamins and minerals were bought from Morgan Co. in Cairo, Egypt.

The other chemicals used in the experiment, including formalin, ethanol, and EDTA were supplied from El-Nasr Pharmaceutical Chemicals, El-Ameriea, Cairo, Egypt.

2.2 Methods

2.2.1 Preparation of aqueous extract of Aloe vera:

Aloe vera L. whole fresh leaves were recognized by a botanist at Menoufia University's Agriculture College in Egypt after being removed from a locally available plant. After thoroughly washing whole aloe vera leaves in lots of water, the gel was removed, combined with distilled water, and shaken. After that, the preparation was filtered out and combined with yogurt to create Formula 1 [18].

2.2.2 Preparation of aqueous pomegranate peel extract

The pomegranate peel was removed and stored at 4 °C in a refrigerator. After being cleaned, the peels were cut into tiny pieces and processed in a grinder. The extract was then made in water with a solvent ratio of 1:50 and heated to 80 °C for 30 minutes [19]. In the rotary vacuum evaporator, the extracted material was cooled, filtered, and concentrated at 60°C and 20 rpm until the volume was reduced to 4/5 of the total extract [20]. The concentrated extract was kept in a dark, airtight plastic bottle at -18 °C. [21].

2.2.3 Preparation of yogurt

At Menoufia University's Faculty of Agriculture, yogurt was produced. Dairy Laboratory uses plain Dannon® yogurt with Lactobacillus and Bifidobacteria species at pH 4.5 to 4.8 as a fermentation starter. 20% of the cultured yogurt was solid..

2.2.4 Induction of gastric ulcer:

For two hours, the rats in the experimental and positive control groups were given a single oral dosage of ethyl alcohol (10 ml/kg body weight) to cause stomach ulcers [22].

2.2.5 Assessment of gastric mucosal injury

Histological sections were created by fixing stomach tissues in 10% formalin for a whole day in order to confirm the presence of peptic ulcers. Histological analysis of hemorrhagic regions in ulcer cases revealed that the stomach pits and surface epithelium were either absent or severely damaged, which led to the emergence of subepithelial capillaries. [23].

2.2.6 Experimental design

For seven days, 35 mature male white albino rats of the Sprague Dawley strain, weighing 150g±5 g. The rats were housed in wire cages that were cylindrical and had wire bottoms. In order to prevent food from being scattered, the diet was presented in special food containers. Additionally, the rats were given water by projecting a glass tube through the wire cage. The rats were lived in animal quarters with regulated humidity and temperature, a 12-hour light/dark cycle, and unrestricted access to food and water.

The experiment was carried out at the Biology Laboratory, Faculty of Home Economic, Menoufia University, Egypt.

For seven days, all rats were fed the basal diet, as described by the American Institute of Nutrition (AIN)[24].

Rats were divided into two main groups, the first group, known as the negative control group, consisted of seven normal rats who did not receive any intervention; and they were fed on basal diet and oral dose (5 ml/kg) of distilled water only.

The Second group (n=28 rat) is the gastric ulcer-induced rats that were randomly divided into four equal subgroups (7 rats):

Positive control group (PCG): include ulcerative gastric rats that were infected with gastric ulcer and they were fed on basal diet and oral dose (5 ml/kg) of distilled water only.

Aloe vera group (AvG): include ulcerative gastric rats that were treated with oral administration of formula 1 (Aloevera 200 mg/kg [18], in yogurt at dose 5 ml/kg [25].

Pomegranate group (PmG): include ulcerative gastric rats that were treated with oral administration of formula 2 (Pomegranate peel extract (50 mg/kg per day in yogurt at dose 5 ml/kg) [26].

Combined group (APG): ulcerative gastric rats were treated with oral administration of formula 3 (Aloe vera and pomegranate (1:1) in yogurt.

At the beginning of experiment, blood samples of the rats were tested to get the initial values before the dietary intervention.

At the end of experimental periods the rats from each group were fasted for 12 hours, then slaughtered and blood samples were collected from hepatic portal vein into a dry clean centrifuge tube. Blood samples were centrifuged at (4000 rpm) for ten minutes to separate blood serum, then kept in deep freezer till using.

2.2.7 Diets

Standard diet: The basal diet was made in accordance with AIN. [24].

2.2.8 Biological Evaluation:

Body weight gain (BWG), food intake (FI), and feed efficiency ratio (FER):

Throughout the trial, body weight was measured once a week and net food consumption was measured every day. The following is how feed efficiency ratios (FER) were calculated using net food intake and acquired body weight:

$$\text{FER \%} = \frac{\text{Body weight gain (g)}}{\text{Food intake (g)}} \times 100$$

2.2.9 Biochemical analysis

Using the techniques of A.O.A.C. [27], a sample of the prepared pomegranate and aleovera was taken to estimate its chemical

composition (moisture, protein, fat, ash, total sugars, some sugars, and polysaccharide).

The method used for the determination of total phenols using Folin-Ciocalteau reagent was adapted from [28]. To ascertain the sample's total flavonoid content, the aluminum chloride colorimetric method was employed, following the procedure outlined by Miliauskas et al. [29]

Assessment of antioxidant status: erythrocyte red blood cells' levels of Super Oxide Dismutase (SOD), Malondialdehyde (MDA), and Catalase (CAT) were measured using a spectrophotometer approach in accordance with Stroev and Makarova [30].

Collection of gastric juice: Rats will be fasted for 12 hours at the end of the experiment, and all rats were sedated with diethyl ether two hours after ethyl alcohol is administered. Then the gastric juice was collected and centrifuged to examine the characteristics of gastric secretion.

Each stomach's contents were moved into a tube from the centrifuge tubes, and the tubes' contents were spun for ten minutes at 1000 rpm. The floating liquid was then moved to measuring cylinders to determine the volume. To measure the pH, one milliliter of gastric juice was collected, and a digital pH meter was used to take a direct reading [31].

Evaluation of Stomach ulceration degree

The ulcer score, which is determined by dividing the total number of ulcers in each group by the number of rats in that group, was used to express the degree of ulceration [32].The ulcer score was multiplied by 100 to determine the ulcer index (UI) [33], The preventative index was computed using the method of [34], and the ulceration (%) was computed by dividing the number of animals with ulcers by the total number of animals and multiplying by 100 [35]. The percentage protection was measured in equation (2): % Protection = (Ulcer Index) control − (Ulcer Index) test / (Ulcer Index) control × 100. The severity of ulcers was classified as described, Normal coloured stomach: 0; Red colouration : 0.5; Spot ulcer: 1; Hemorrhagic streaks: 1.5; Ulcers ≥ 3 but ≤ 5; Severe ulcers >5 : 3.

The histological characteristics of gastric lesions were examined under a microscope by preserving stomach tissues in 10% formalin for a whole day. Haematoxylin and eosin dye is used to stain the paraffin-made specimens after they have been sectioned to a size of 3-5 µm. An optical microscope was used to examine the histological sections.

2.2.10 Statistical Analysis:

The mean ± SD was used to tabulate the results. A statistical analysis system was used to do an analysis of variance (ANOVA) on the experimental data for a completely randomized design [36]. To ascertain mean differences at the 5% level, Duncan's multiple range tests were employed.

2.2.11 Ethical approval

All the experimental and animal care protocols were ethically approved by the Institutional Animal Care and Use Committee (IACUC) at Menoufia University, Sheibin El-Kom, Egypt (Approval No. MUFHE/S/NFS/15/24). All biological experiments were performed in compliance with the policies of the IACUC for the use and care of laboratory animals. Top of Form

3. Results and Discussion

The study had shown the potency of dietary intervention with alovera and pomegranate extract as antiulcer formula of induced rats with gastric ulcer.

Data in Table (1) showed the effect of pomegranate and alovera fortified with yogurt on body weight gain, feed intake, and feed efficiency ratio of the experimented groups. There was a significant ($p<0.05$) reduction in BWG in the positive control group (58.00±3.92 g) compared with the negative control group (116.87±4.61 g) and treated groups. This was in line with previous findings by Tahoon and El Sheikh [37] who reported that FI, BWG and

FER were decreased in ulcer peptic groups than normal control.

These results were consistent with those of Malik et al [38], who reported that weight loss may be a consequence of gastric ulcer pain, which intensifies two to three hours after a meal. Due to the enlargement of the ulcerated area, peptic ulcers can also produce a blockage in the digestive tract, resulting in weight loss, frequent vomiting, and an early sense of satiety. Peptic ulcers could increase the danger of internal bleeding, and until the stomach or small intestinal lining was pierced, open sores may continue to grow, putting the patient at risk for peritonitis [Ministry of Health, 2025]. This can lead to malnutrition, especially if there is stenosis, which might prevent normal food intake. Thus, anemia, melena, hematemesis, or weight loss were all signs of peptic ulcer problems in any patient. While the ulcerative groups that fortified with combined pomegranate and alovera fortified with yogurt had the best ameliorative effect, followed by AVG, then PMG by means values of 106.62±3.73, 98.75±1.83, and 80.87±2.79 g/28day/r, respectively. These results were in harmony, with those obtained by Ismael et al [39] and Nna et al[40] who indicated that aloe vera suspension gel consumption was associated with higher body weight.

The significant improvement in body weight observed when taking aloe vera extract, that could be attributed to the good influence on the nature of the small intestinal flora. The ability of the aloe vera plant to improve digestion efficiency due to its enzymes content, such as carboxy-peptidase, catalase, cellulose, and peroxidase, may be responsible for the increase in live body weight. Additionally, this plant contains many vitamins, including vitamins (A, C, and E), group B vitamins, folic acid, and choline. B vitamins play a role in amino acid metabolism [41]. Similar results were obtained by Aziz and Al-badry[42] who reported that rats which received pomegranate peel extract at different concentrations gained significantly more weight than those who had stomach ulcers.

Moreover, the reason for the significant weight gain could be attributed to the antioxidant properties of pomegranate peels, which enhance the animals' eating abilities. This is consistent with the findings of Li et al. [43], who demonstrated that the peels contain antioxidants in the form of multiphenol compounds and tannins.

Pomegranate peels are a good therapy option because they include antioxidants and vitamins. This is in line with the findings of Kulkarni et al. [44], who discovered that pomegranate peels had high quantities of vitamin B5 and vitamin C. Another factor contributing to this increase is the part that plant components play in lowering lipid peroxidation brought on by stomach ulcers. Heeb et al. [45] discovered that indomethacin causes lipid peroxidation and the production of free radicals. This is comparable to the findings of Larrosa et al. [46], who reported that the tannins in pomegranate peels had the ability to scavenge free radicals and stop lipid peroxidation.

Consistent with the conclusions stated by Haque et al. [47] , who observed that using yogurt as a dietary supplement can increase body weight gain without changing blood triglyceride and cholesterol levels. This outcome supported the conclusions of Sultana et al. [48], who showed that using 5g yogurt as a nutritional supplement boosted weight gain as opposed to the control group. The activity of probiotic bacteria is one of the reasons why living organisms gain weight. According to earlier studies, the utilization of nutrients and their absorption by absorptive cells are what lead to the increase in digestive efficiency [49]. According to earlier research, the presence of probiotic bacteria increased the morphometric surface of intestinal villi and the activity of digesting enzymes. Increased digestibility, nutrient absorption, and the development and enlargement of new tissues are all impacted by this physiological state

[50]. Other researchers have also published the findings of the same study [51].

Adriani et al. [50] have also documented an increase in the number and width of the jejunal villi from diet plus probiotics that affects the broilers' ultimate body weight.

Furthermore, several researchers have highlighted that boosting the population of microorganisms that are advantageous to cattle would stop the growth of pathogenic microbes in the food's digestive tract [52].

Regarding FI, the most observative findings were that APG increased the amount of food intake that reached the same intake of the negative control group without significant difference, and PMG did not change more than APG while differing significantly ($p<0.05$) from NCG, being 16.86±0.77, 17.67±0.65, and 15.94±1.00 g/day, respectively. While AVG ate more, it consumed the least amount of food (14.93±0.99 g/day) compared to the other fortified groups, with no discernible difference. Our findings are in line with those of Setyaningrum et al. [53]and Khan et al. [54], Who discovered that consuming yogurt as a dietary aid elevated feed intake (FI), FER, and live weight gain of broiler chickens.

For FER, the treated group with combined pomegranate and alovera fortified with yogurt had a better feed efficiency influence without a significant difference from NCG; the values were 0.22±0.013 and 0.23±0.012, respectively. On the other hand, the other experimented groups supported by each type separately improved but differed significantly ($p<0.05$) when compared with PCG by means values of 0.20±0.018, 0.18±0.009, and 0.13±0.017 for AVG, PMG and PCG, respectively.

By enhancing digestion and encouraging feelings of fullness, aloe vera juice may help with weight management. It can help control hunger, lessen cravings, and promote a healthy metabolism when consumed on an empty stomach. Polysaccharides found in aloe vera aid in the intestines' absorption of nutrients. The bioavailability of vitamins, minerals, and other nutrients consumed throughout the day may be improved by consuming aloe juice on an empty stomach [54].

According to the study's findings, taking supplements of pomegranate peel considerably lowers body mass index, weight, waist circumference, and fat mass index. In line with our findings, a 2019 study on rats fed a high-fat diet discovered that the group that received cake with 15% pomegranate peel powder gained less weight than other groups [56]. Nonetheless, data indicates that pomegranate peel's appetite-suppressing benefits are more pronounced when a high-fat diet is followed. Pomegranate peel has been shown in animal tests to have a favorable effect on hunger by influencing leptin. Pomegranate peel powder administration significantly reduced leptin levels in another study conducted by Soliman et al.(57] on overweight mice. Pomegranate peel's ability to suppress appetite may account for the treatment group's (PMG) absence of noticeable weight reduction during the research and their lower level of diet acceptance compared to the Alo vera intervention group.

The data presented in Table 2 illustrate the effect of yogurt-fortified pomegranate and alovera on complete blood cells (CBC). Prior to the onset of the experiment, all infected groups had significantly ($p\leq0.05$) lower RBC than NCC. Compared to the treated groups and the positive control group (3.46±0.44 10-6/cm m), the RBCS count of the rats in the APG (7.03±0.59) was significantly higher ($p\leq0.05$) following the dietary intervention. However, compared to all experimental groups, the negative control group had a significantly higher value (8.56±0.59 106/cm m). There was no significant difference ($p<0.05$) in the count of RBCS between AVG and PMG.

Table (1). Effect of dietary intervention with pomegranate and alovera fortified in yogurt on BWG, FI and FER of induced rats with peptic ulcer

Groups	BWG (g/28day/r)	FI (g/day)	FER (%)
	Mean±SD	Mean±SD	Mean±SD
NCG	116.87a±4.61	17.67a±0.65	0.23a±0.012
PCG	58.00 e±3.92	12.28c±1.02	0.13d±0.017
AVG	98.75c±1.83	14.93b±0.99	0.20c±0.018
PMG	80.87 d±2.79	15.94b±1.00	0.18b±0.009
APG	106.62b±3.73	16.86ab±0.77	0.22a±0.013
LSD	7.841	1.652	0.013

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant ; BWG: Body weight gain; FI: Food Intake; FER: Feed Efficiency Ratio; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

As for blood hemoglobin (HB) values, before dietary intervention, the induced rats with peptic ulcers showed a significant decrease compared to the negative control group (15.40±0.46 g/dl). while it had significantly increased among treated groups when compared to the positive control group (7.37±0.65) after dietary intervention, with the highest significant ($p<0.05$) values in APG (12.25±0.70 g/dl).

Moreover, the highest hematocrit (HCT) value (41.37±2.39) was found in rats of the APG which nearly to the negative control group 43.24a±1.89 but differed significantly ($p<0.05$), and the lowest HCT (29.94±1.33) was found in rats of the positive control group. There were no significant differences between AVG and PMG in their effect on HCT, which were 38.80±1.58 and 38.02±0.77%, respectively.

In line with the findings of Gopinathan [58], and Rajendran et al. [59], who noted that supplementing with aloe vera gel improved counts of red blood cells and enhanced blood hemoglobin and oxygen carrying capacity. Niewiadomska et al. [60]and Mahmoud et al. [61] showed similar results, stating that PPE co-administration enhanced RBC count, hemoglobin content, and hematocrit percentage. This demonstrates that PPE can help for enhanceing RBC production, maturation, and survival. PPE compounds may stimulate erythropoiesis and protect RBCs from oxidative stress.

Table (2). Effect of dietary intervention with pomegranate and alovera fortified in yogurt on RBCs, Hgb and Hct of induced rats with peptic ulcer.

Groups	RBCs (106/cm m)			Hgb(g/dl)			Hct(%)		
	PRE	Post	Sig	Pre	Post	Sig	Pre	Post	Sig
NGG	9.62a±0.81	8.56a±0.59	0.015	15.40a±0.46	15.51a±0.43	0.594 ^{NS}	41.9a±1.04	43.2a±1.89	0.108 ^{NS}
PGG	3.50b±0.45	3.46d±0.44	0.883^{NS}	7.37b±0.65	7.41d±0.43	0.914 ^{NS}	29.4bc±1.69	29.9d±1.33	0.525 ^{NS}
AVG	3.58b±0.43	6.00c±0.58	0.000	7.27b±0.60	10.9c±0.48	0.000	29.6bc±0.93	38.8c±1.58	0.000
PMG	3.87b±0.37	5.62c±0.66	0.000	7.53b±0.68	10.0c±0.31	0.000	30.1b±0.67	38.0c±0.77	0.000
APG	3.69b±0.35	7.03b±0.59	0.000	7.02b±0.48	12.25b±0.70	0.000	28.3c±1.48	41.3b±2.39	0.000
LSD	0.381	0.402	--	0.522	2.553	--	1.466	1.850	--

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant; RBCS: Red Blood Cell Count; Hgb: Hemoglobin; Hct:Hematocrit; NGG: negative control (-ve) group; PCG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

The results are consistent with those of Al-Hadidi [62] who showed that probiotics can increase the number of red blood cells and aid in their return to normal levels via a variety of methods. The drop in pH ascribed to the fermentation processes induced by probiotics is a crucial factor among them. Additionally, as noted by Abudabos et al. [63], the synthesis of vitamin B-complex by probiotics has been found to be a major factor in the

enhancement of red blood cell quality, control of hemoglobin concentration, and regulation of blood cell size. This support from earlier studies highlights the positive effects of probiotics on hematological markers. Also, this outcome is consistent with the findings of Marcia et al. [64], who revealed that yogurt therapy improved hematological markers such as HG and RBC over 4 weeks. Hoppe et al. [65] reported that in people with an increased need for iron supplementation of L. plantarum 299v together with a meal enhances the bioavailability of iron.

The data recorded in Table (3) reflected the effect of oral administration of yogurt-fortified pomegranate and alovera on MCV, MCH, and MCHC. At the beginning of the experiment, there were non-significant differences observed in the level of MCV among all ulcerative groups and each other, but there was a significant increase in ulcerative gastric rats compared with the positive control group (51.32±0.84 fl). At the end of the experimental periods, APG had the greatest ameliorative benefit when compared to the negative control group after AVG, followed by PMG by means values of 69.21±1.10, 79.90±2.83, 65.47±2.09, and 63.30±1.94 fl, respectively.

As well, the mean corpuscular hemoglobin (MCH) count was 28.36±1.77 pg in healthy control animals, and it drastically decreased to 16.84±.88 pg in the positive control animal. But the other groups of animals that were treated significantly enhanced the MCH count to 24.44±2.05, 23.48±1.56 and 21.84±0.88 pg for APG, AVG and PMG, respectively, showing no significant distinction between APG and AVG. These findings concurred with those of earlier research. Salama et al. [66] demonstrated that oral supplementation of 0.50g\ kg aloe vera gel significantly increase mean corpuscular hemoglobin (MCH), mean corpuscular volume (MCV) and MCHC.

In the same table, the mean corpuscular hemoglobin concentration (MCHC) count was lower in the ulcerative groups than in the in the negative control group (33.62±1.83 g/dl) before dietary intervention. While the ulcerative group that fortified with combined pomegranate and alovera with yogurt showed a higher count of MCHC (30.02±0.86 g/dl) than the positive control group (21.38±.86 g/dl) and the other fortified group, there was no significant difference ($p \leq 0.05$) between AVG and PMG.

There are strong research data from Zakrzewska et al. [67] who mentioned that some probiotics, such as Lactobacillus acidophilus and Bifidobacterium longum improve iron absorption and influence the course of anemia, this may be due to the ability of prebiotics to lower the pH of the colon, increasing the reduction of Fe^{3+} to Fe^{2+}, as well as the fermentation process that enhances SCFA production, which may contribute to increased colon absorption area [68]. The explanation for these findings could be that probiotics help lessen oxidative stress, which supports the health and function of red blood cells. To reduce oxidative damage, they do this by producing antioxidants such as glutathione, folate, and short-chain fatty acids and by strengthening endogenous antioxidant enzymes that neutralize reactive oxygen species. RBC, HCT, MCH, and MCHC levels are stabilized by probiotics through the reduction of oxidative stress [69]. These results also align with those of Koker et al. [70], who found that probiotic supplementation during the first 30 days of iron replacement therapy (IRT) improved blood iron indicators and Hb levels in individuals with iron deficiency anemia (IDA).

Furthermore, probiotics may improve the absorption of vital nutrients like vitamin B12 and folate, which are necessary for the synthesis of red blood cells and the maintenance of MCH and MCHC levels, by promoting gut health. This enhanced nutritional absorption may support hematological health and mood stabilization by promoting optimal RBC, HCT, MCH, and MCHC levels [71].

Table (3). Effect of dietary intervention with pomegranate and alovera fortified in yogurt on MCV, MCH and MCHC of induced rats with peptic ulcer.

Groups	MCV(fl)			MCH(pg)			MCHC(g/dl)		
	Pre	Post	Sig	Pre	Post	Sig	Pre	Post	Sig
NGG	81.57a±1.68	79.90a±2.83	0.043	29.25a±1.67	28.36a±1.77	0.261^{NS}	33.62a±1.83	35.29a±.83	0.073^{NS}
PGG	51.00b±1.01	51.32e±0.84	0.565^{NS}	16.88b±0.84	16.84d±.88	0.917^{NS}	21.69b±0.54	21.38d±.86	0.142^{NS}
AVG	51.67b±0.99	65.47c±2.09	0.000	16.77b±0.77	23.48b±1.56	0.000	21.54b±0.81	28.7c±1.24	0.000
PMG	51.50b±1.19	63.30d±1.94	0.000	17.43b±0.87	21.84c±0.88	0.000	21.29b±0.80	28.1c±0.91	0.000
APG	51.43b±1.16	69.21b±1.10	0.000	16.86b±0.86	24.44b±2.05	0.000	21.18b±0.72	30.0b±0.86	0.000
LSD	2.170	2.069		0.109	1.107		2.750	1.041	

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant ; MCV:Mean Corpuscular Volume;MCH:Mean corpuscular; MCHC:Mean Corpuscular Hemoglobin concentration; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

The results in Table 4 indicated the effect of yogurt fortified with pomegranate and alovera on the platelet count, white blood cells, and percentage of eosinophils. Prior to the oneest, there was a significant (p<0.05) reduction in the platelet count in the ulcerative groups compared with the negative control group (837.85±5.32*103/ul). While it had significantly increased (p≤0.05) among all treated rat groups when compared to the negative control group following nutritional therapy, with the highest significant (p<0.05) values in the mixed group (790.00b±5.75*103/ul).

Within the same table, the white blood cell count was higher in the disease-control rat than healthy control animals (6.17 ± 0.82*103/ul). But the pre-treated groups with nutritional therapy brought down the WBC in their groups. The amount of WBC in the combined group (8.55 ± 0.87*103/ul) was almost comparable to the healthy control group. There was no significant difference (p≤0.05) in AVG and PMG with values of 9.50 ±0.81 and 9.78 ±0.58*103/ul, respectively. Additionally concurring with these conclusions, El Bohi et al. [88] reported that PPE can impact leukocyte survival, differentiation, or generation, assisting in the maintenance of a well-balanced immune response overall. Moreover, the results obtained today also aligned with those released by Darabighane et al. [89] who claimed that adding aloe vera gel to broiler diet increased the number of white blood cells in the birds overall. Also, this outcome is consistent with the findings of Marcia et al. [64], who revealed that yogurt therapy improved hematological markers such as WBC over 4 weeks.

Concerning the EOS, at the start of the trial, there were no significant variations (p≤0.05) in the percentage of eosinophils across all ulcerative groups and each other. Although there was a substantial increase (p≤0.05) in ulcerative gastric rats compared to the negative control group (2.45±0.53%). After the trial has ended, The value of the combined group (3.23 ± 0.58%) was the lost ,almost equal to that of the negative control group when compared to the positive control group (5.80 ± 0.35%) and the other fortified group. There was no significant difference (p≤0.05) between APG and AVG or between AVG and PMG. Although there was a significant difference (p<0.05) between APG and PMG. These outcomes also concurred with Hasan et al.'s [90] and Niewiadomska et al. [60] recorded data, which revealed that the group that received treatment with pomegranate peel aqueous extract exhibited a noteworthy rise in eosinophil count when compared to other groups.

Table (4). Effect of dietary intervention with pomegranate and alovera fortified in yogurt on PLT, WBCs and EOS of induced peptic ulcer rats.

Groups	PLT(*10^3/ul)			WBCs(*10^3/ul)			EOS(%)		
	PRE	Post	Sig	Pre	Post	Sig	Pre	Post	Sig
NGG	837.8a±5.32	828.7a±6.62	0.001	6.17c±0.82	5.74c±0.80	0.291^{NS}	2.45a±0.53	2.82d±0.54	0.353^{NS}
PGG	677.4c±5.23	688.8e±4.85	0.011	15.39a±0.55	15.70a±0.63	0.303^{NS}	5.78b±0.46	5.80a±0.35	0.950^{NS}
AVG	687.4b±4.65	774.4c±4.03	0.000	14.7ab±0.79	9.50b±0.81	0.000	6.16b±0.49	3.5bc±0.35	0.000
PMG	667.7d±5.09	766.5d±4.13	0.000	14.45b±0.63	9.78b±0.58	0.000	5.91b±0.51	3.89b±0.31	0.000
APG	659.2e±5.09	790.0b±5.75	0.000	15.0ab±0.97	8.55c±0.87	0.000	6.07b±0.42	3.2cd±0.58	0.000
LSD	2.573	7.521		0.910	0.932		0.438	0.566	

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant ; PLT: Platelet Count; WBC: White Blood Cells; EOS: Eosinophils; ; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

Table (5) exhibits the effect of Dietary Intervention with Pomegranate and Aleo vera on antioxidant enzymes and oxidative stress indicators of rats with peptic ulcer. the antioxidant as measured by Superoxid dismutase (SOD) and Catalase (CAT) and the oxidative stress were measured by Malondialdehyde (MDA).in compaision to the negative control group(31.09±0.77u/ml), the rats which received ethyle alchol shown aconsiderable decrease in SOD level before the food intervention. Also, in agreement with these findings, Alotaibi et al. [72] reported that when rats were given ethanol, there was a decrease in SOD level.

More significantly rise (p≤0.05) in SOD level among treated groups when compared to the positive control group(10.46±0.35u/ml) following a nutritional intervention. the best results are existed in the combined group (APG), where it recorded values(25.03±0.55u/ml). The current findings were also consistent with those published by Abozahra et al. [73] and Abdel Moneim [74], who discovered that treatment with PPE in diets had a large increase in SOD activity and a considerable decrease in MDA in a dose-dependent manner. These results could be explained by the polyphenolic chemicals (catechin, naproxene, ellagic acid, gallic acid, cholinergic acid, and vanillin) present in PPE. Because of their antioxidant qualities, these substances lessen oxidative stress by reducing lipid peroxidation, hydrogen peroxide, and superoxide anion levels. They also have potent anti-free radical properties.[75] and [76]. Also, Ali et al. [77], who reported that probiotics can increase SOD levels because probiotics can reduce oxidative stress and inflammation.

Additionally, there was a high statistical difference(p<0.5) between all treatment groupings and the positive control group(0.69±0.15ng-ml) in CAT activity. The highest CAT activity (10.56±1.35). was seen in the negative control group followed by combined group (6.65±1.04ng-ml). no statistical significance was found between AVG and PMG with values of 4.72±0.92 and 3.56±0.80ng-ml, respectively. These results were in line with those of previous studies Hussein et al[78] who reported that aloe vera decreased oxidative stress as seen by higher gastric CAT levels and decreased stomach MDA). Aloe vera has a strong antioxidant impact because of its ability to scavenge free radicals and chelate metals [79]. It is also brought on by a high concentration of enzymes (catalase, superoxide dismutase, and glutathione peroxidase), phenolic antioxidants (chromones, coumarins, saponins, flavonoids, and tannins), and antioxidant-containing vitamins (A, E, and C). The OH group preserves the thiol content of tissues and inhibits the oxidation of cellular proteins by preventing the -SH group from being broken down oxidatively [80]. These results corroborate Singh et al. [81] observations, which stated that Pomegranate peel extract boosted catalase (CAT), and superoxide dismutase (SOD) in mice while decreasing MDA levels. As a result, using pomegranate peel in decoctions (concentrated aqueous heated extract) may

increase the concentration of phenolic components, dietary fiber, polysaccharides, vitamins, and minerals [82].

When examined in terms of oxidative stress parameters, MDA levels had significantly increased (p≤0.05) in the Rats with stomach ulcers when compared to the normal group (1.17±0.23 nmol-ml) before the study began. This was consistent with Chams and Eissa[83], Abdel-Kawi et al.[84] and Paulrayer et al. [85], finding that ethanol administration in rats resulted in an increase in MDA concentration There is evidence linking oxidative stress to the pathogenesis of GU. It arises from an imbalance between ROS and antioxidants, which causes GU. By causing lipid peroxidation and the stomach mucosa's antioxidant reserves to be depleted, alcohol causes oxidative stress. Lipid peroxidation culminates in MDA, which is widely employed as a reliable marker of lipid peroxidation [86].

While there were significantly decrease among treated groups when compared to the positive control group (9.36±1.22 nmol-ml) after dietary intervention. The Malondialdehyde activity of the APG was the lower than those of both AVG and PMG by means values of 1.54±0.53, 2.77±0.50 and 3.79±1.11 nmol-ml), respectively. These results are consistent with those obtained by Ejtahed et al. [87], who found that there was a significant drop in MDA levels in the probiotic treated group following intake of probiotic yogurt.

Table (5). Effect of dietary intervention with pomegranate and alovera fortified in yogurt on antioxidant indicators of induced rats with peptic ulcer

Groups	SOD (u/ml)			CAT(ng-ml)			MDA(nmol-ml)		
	PRE	Post	Sig	Pre	Post	Sig	Pre	Post	Sig
NGG	31.09a±0.77	30.47a±1.81	0.360^{NS}	11.73a±0.87	10.56a±1.35	0.148^{NS}	1.17d±0.23	0.71e±0.99	0.001
PGG	10.04b±0.90	10.46e±0.35	0.230^{NS}	0.58b±0.16	0.69d±0.15	0.125^{NS}	11.0bc±1.55	9.36a±1.22	0.022
AVG	9.89b±0.72	22.56c±0.54	0.000	0.44b±0.13	4.72c±0.92	0.000	10.68b±0.98	2.77c±0.50	0.000
PMG	10.70b±0.85	20.06d±0.61	0.000	0.67b±0.12	3.56c±0.80	0.000	9.67b±1.44	3.79b±1.11	0.000
APG	9.80b±0.87	25.03b±0.55	0.000	0.33b±0.15	6.65b±1.04	0.000	14.54a±1.72	1.54d±0.53	0.000
LSD	1.034	1.471	--	0.432	1.287	--	1.102	0.772	--

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant; SOD: Superoxide dismutase; MDA : Malondialdehyde ; CAT: Catalase ; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

The statistical data presented in Table 6 shows that the effect of pomegranate and aloe vera fortified with yogurt on the volume and PH values of gastric juice in normal rats and rats with stomach ulcers. A significant reduction (p≤0.05) in the volume of gastric juice (2.40±0.47 ml/100g) was observed in the combined group, almost to the negative control group (2.53 ml/100 g) when compared to the positive control group (4.82±0.22 ml/100 g). Followed by AVG and PMG by means values of 3.50±0.44 and 4.02 ±0.34 ml/100 g, respectively. The result of the study came in accordance with Piracha et al.[91] and Alimi et al.[92]who reported that the treatment groups that received the extract of Punica granatum (pomegranate) peel demonstrated a significant decrease in gastric juice volume, and increase in gastric juice ph. Furthermore, it has been discovered that pomegranate peel extracts raise PGE2 and NO levels, reduce oxidative stress, and prevent the release of pro-inflammatory cytokines. The stomach H+, K+-ATPase enzyme, which is crucial for controlling gastric ulcers and lowering the generation of gastric acid, was likewise effectively inhibited by the extracts.

Regarding PH, when comparing the PH value of gastric juice to that of normal rats, the ulcerated rats without treatment experienced the highest decrease, which was recorded at 1.37±0.12 mEQ/L. These results are in harmony with those obtained by Sanpinit et al. [93] and Rahman et al. [94] who indicated that when ethanol is injected, the pH of the gastric juice decreases, gastric mucus

synthesis decreases, and inflammatory cells infiltrate the gastrointestinal mucosa because of injury. Conversely, all treated groups of gastric ulcer rats significantly increased (p≤0.05) the PH value compared to the positive control group, and the combined group showed the highest significant increase (3.90 ±0.33 m EQ/L) with no significant difference (p≤0.05) from the negative control group, which recorded (4.03 ±0.35 m EQ/L). These outcomes supported the findings of previous studies Retiu et al. [95] who reported that aloe vera treatment markedly increased stomach pH because it prevented the release of acid). Aloe vera increases gastrointestinal pH, promoting normal enzyme activity and iron and calcium absorption.The plant's lectin content could be the reason behind aloe vera's inhibitory action [86]. Lectins prevent parietal cells from absorbing aminopyrine. As a result, reducing stomach acid is the direct outcome of influencing acid-secreting cells [96]. Retiu et al. [95] showed that gel aloe vera can be used to maintain a normal gastric pH of 1-2 to 4-5 and expedite the reduction of stomach acid secretion (HCl generation). Reduced HCl in the stomach result in a reduction in gastritis discomfort. The primary cause of aloe's laxative effect is the presence of aloin A and B, 1, 8-dihydroxyanthracene glycosides. The alkaline content of aloe vera aids in balancing and reducing excess stomach acid.

The data presented in table 7 elucidated the effect of oral administration of yogurt-fortified pomegranate and alovera on the number of ulcers, ulcer score, and ulcer index of stomach ulcer rats. There is a reduction in the average ulcer number in all treaded rat groups compared to the positive control group, which has an average ulcer number of 6.37±0.51, and the lowest average (1.12±0.99) was found in the combined group. There was no significant difference (p≤0.05) in the average number of ulcers counts between AVG and PMG by means values of 2.87±0.83 and 3.00±1.30, respectively. Therefore, the ulcer score, The ulcerated control group had the highest value in the ulcer score (10.03±1.05) compared with all treated groups, whereas the ulcer score of the APG was lower than those of both AVG and PMG with values of 3.34±0.45, 5.20±0.54, and 6.93±0.54, respectively.

Table (6). Effect of dietary intervention with pomegranate and alovera fortified with yogurt on Volume and PH of gastric juice of induced peptic ulcer rats.

	Gastric Vol(ml/100g)		PH(m EQ/L)	
Groups	M±SD	Sig	M±SD	Sig
NGG	2.53d±0.43	0.000	4.03a±0.35	0.000
PGG	4.82a±0.22		1.37d±0.12	
AVG	3.50c±0.44		2.95b±0.34	
PMG	4.02b±0.34		1.94c±0.23	
APG	2.40d±0.47		3.90a±0.33	
LSD	0.410		0.527	

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant;GV:Volum of gastric acid;PH:Potential of Hydrogen; ; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

When estimating the ulcer index, the disease control group's mean index was severe (1620.81±433.29), but it decreased in the other animal groups. This agreed with the reports of Karampour et al. [97] and Bakry et al. [98] that the ulcer index of experimental animals increase after ethanol administration. The APG (359.71±44.14) showed the maximum inhibition of the ulcer index, with no discernible change from the AVG. While APG differed considerably (p<0.05) from PMG by mean values of 724.18±15.17, 542.15±21.68, and 359.71±44.14 for PMG, AVG, and APG, respectively, PMG did not vary more than AVG.

The result of the study came in accordance with Piracha et al. [91] and Chauhan et al. [99] and Ajaikumar, et al.[100] who reported that the treatment groups that received the extract of Punica granatum (pomegranate) peel demonstrated a significant decrease in ulcer index. These findings concurred with earlier research Silla et al. [101], which revealed that PPE dramatically slowed the growth of

bacteria and successfully decreased oxidative stress and inflammation in human gingival epithelial cells (HGECs). Pomegranate's anti-inflammatory and antioxidant qualities were credited in these studies with its protective qualities. The strong anti-inflammatory and antioxidant properties of PPE are attributed to its natural polyphenols, which include flavonoids and tannins. In fact, these benefits were supported by the rise in TT that coincides with the drop in TNF- αlevels in the pomegranate peel-treated groups. PPE's protective qualities are probably due to the bioactive substances found in it, such as (E)- 9-octadecenoic acid methyl ester, methyl 9-cis, 11-transoctadecadienoate, and other fatty acids. Strong antioxidant and anti-inflammatory properties of these substances may mitigate oxidative stress and inflammatory reactions [61].

These outcomes aligned with the findings of Mohamed et al. [102], who reported that treatment with aloe vera gel significantly decreased UI. The active components of aloe vera gel, such as tannins, saponins, and flavonoids, may be responsible for its cyto-protective properties [103]., Although aloe vera gel has been shown in numerous studies to be an effective medium for wound healing it is still unknown what precise mechanism causes and speeds up wound healing (104). Aloeemodin, ßsitosterol, and ß-sitosterol glucoside are its three angiogenic constituents [105]. Plasminogen activator can be strongly induced by ß-sitosterol, which has a direct impact by raising the expression of the VEGF gene, which results in a rise in growth in the number of new blood vessels and the proliferation of endothelial cells [106].

Aloe vera's tannins, saponins, and flavonoids, which are its active ingredients, may be the reason for its capacity to heal wounds. Tannins and other polyphenolic compounds have been used as GU treatments because of their astringent qualities. To form a barrier over the injured tissues, they interact with tissue proteins in the GU and precipitate microproteins at the ulcer site. In addition to shielding the underlying mucosa from harsh stimuli and preventing stomach secretion, the barrier also speeds up the healing process [107]. Auxin and gibberellin, which function as growth hormones and promote cell growth and regeneration to aid in wound healing, are also present in its gel [108]

Pomegranate peel powder (10%) has been introduced as an efficient and cost-effective antiulcer compound by Mohamed and Mabrok [109], who found that it has an antiulcer effect in tested rats by reducing the expression of inflammation markers and regulating NO, which increases mucus secretion.

Probiotics may strengthen the stomach's mucosal barrier, which guards against stomach acid damaging the stomach lining. Enhancing this barrier can help existing ulcers heal more quickly and have a preventative effect on the emergence of new ulcers.

Probiotics may aid in lowering the production of excess stomach acid, which is a contributing factor to the development of ulcers, according to certain research. Probiotics can help create a more stable environment for the stomach lining by balancing the amounts of stomach acid.

With its high probiotic content, yogurt may help prevent and treat stomach ulcers. Consuming yogurt on a regular basis may help maintain a balanced gut microbiota and provide other healthy nutrients like calcium, proteins, and vitamins that support digestive health in general [110].

According to studies, certain probiotic strains, such Lactobacillus acidophilus, which is present in yogurt, may protect against gastric ulcers by lowering stomach inflammation and inhibiting the growth of dangerous bacteria like H. pylori [111].

Yogurt's probiotics may also help maintain a healthy gut environment, which could aid in the repair of pre-existing ulcers and stop new ones from developing [112].

Table (7). Effect of dietary intervention with pomegranate and alovera fortified with yogurt on Number of Ulcer ,Ulcer Score and Ulcer Index of induced peptic ulcer rats

	No Ulcer		Ulcer Score (US)		Ulcer index (UI)	
Groups	Mean±SD	Sig	Mean±SD	Sig	Mean±SD	Sig
NGG	--		--		--	
PCG	6.37a±0.51		10.03a±1.05		1620.81a±433.29	
AVG	2.87b±0.83	0.000	5.20c±0.54	0.000	542.15bc±21.68	0.000
PMG	3.00b±1.30		6.93b±0.54		724.18b±15.17	
APG	1.12c±0.99		3.34d±0.45		359.71c±44.14	
LSD	1.63		1.137		182.030	

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant;NO Ulcer:number of ulcer;Ulcer S :ulcre score ; UI :ulcer index; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

The data listed in table 8 show the effect of dietary intervention with pomegranate and Aleo vera on the percentage of ulceration, the percentage of the preventive index, and the average severity of ulcers. In ulceration percent, the ulcerated control group had the highest value, which was 82.90±4.70%, followed by the pomegranate group, which was 60.26±3.68%, while the combined group had the least value, which was 35.87±5.89%, followed by the Aloevera group, which was 48.80±2.00%. These findings were consistent with Al-badry and Aziz [42], who reported that providing an aqueous extract of pomegranate peels at various concentrations significantly improved gastrointestinal ulceration in rat models. Also, Extracts from Punica granatum help repair and shield the stomach mucosa from injury [113]. Pomegranate reduced inflammation by inhibiting the production of pro-inflammatory cytokines such as TNF-α, IL-1β, and IL-6, which are known to cause inflammation and damage to stomach tissue [114]. Pomegranate also increased the the synthesis of NO and PGE2, two substances that protect the stomach mucosa [115]. While NO controls blood flow and preserves the gastric mucosal defense systems, PGE2 preserves the integrity of the gastric mucosa and encourages tissue repair [116].

Over and above, there are good values of the protection ratio in all treated rat groups compared to the positive control group (15.92±1.81%), where the combined group had the highest protection rate (60.17±5.24%) compared to the other fortified group, which recorded the following percentages (47.93±3.57 and 37.16±7.29) for AVG and PMG, respectively. This concurs with the findings of Mohamed et al. [102]. Aloe vera treatment significantly improved the protection ratio, which recorded 91% ulcer inhibition. The explanation for these findings could be Aloe vera contains enzymes such bradykinase and carboxypeptidase, which have analgesic and anti-inflammatory properties,according to Ahmed and Hussain [117].

Muhialdin et al.[118] found that the aqueous extract of Punica granatum (pomegranate) peel significantly inhibited the formation of stomach ulcers.The extract's high dose (500mg/kg) inhibited ulcer formation by 84.60%, whereas the low dose (250mg/kg) resulted in a 42.87% inhibition. Omeprazole, the typical medication, only had a 24.50% inhibitory effect.

Also, Alam et al [119] reported that the aqueous methanolic extract of Punica granatum had a significant 87.5% inhibition of ulcerative activity.

In the same table, in comparison to the positive control group, the combined group showed a considerable decrease in the average severity of ulcer scores of 5.57±0.49 and 1.31±0.62, respectively. There were no significant changes among AVG and PMG in their influence on the average severity of ulcers while differing significantly ($p<0.05$) from the NCG. Consistent with the conclusions made by Zedan et al. [120], who observed that Treatment with aloe vera Gel wound healing

accelerated the reepithelization process and prevented any inflammatory reactions in this investigation. This result may be explained by aloe-emodin, which by itself shows an anti-inflammatory response, and by ß-sitosterol, which increases the proliferation of epithelial cells and aids in the reepithelialization of wound healing [121]. According to Chauhan et al.[99] treatment with peel extract from Punica granatum L. decreased the severity of stomach ulceration in rats that was brought on by ethanol (EtOH). PPE's anti-inflammatory properties may primarily stem from ellagitannins' capacity to reduce pro-inflammatory mediators like TNF-α and alter NF-κB signaling, as has already been shown in a number of cell line types [122].

In keeping with the findings of Chauhan et al [123] who noted that Rats administered pomegranate peel extract (Punica granatum L.) demonstrated ulcer-healing and stomach-antiulcer properties .By reducing offensive factors like acid secretion and pepsin activity and increasing defensive factors like mucus secretion and mucosal glycoproteins, the extract demonstrated a strong protective effect against GUs. Additionally, the extract demonstrated antioxidant qualities by raising GSH and CAT activity and decreasing oxidative stress in the stomach mucosa. According to these results, peel extract from Punica granatum L. may be helpful in the treatment and prevention of GUs.

Probiotics protect the gastric mucosal barrier; ii) Prostaglandins, mucus, growth factors, and anti-inflammatory cytokines are upregulated; iii) the ratio of cell proliferation to apoptosis is increased; and iv) angiogenesis is induced. These are some of the general health benefits associated with the cellular and molecular mechanisms of probiotics on various systemic and gastrointestinal disorders that accompany a gastric ulcer [111].

Probiotics have been shown in numerous studies to be effective in treating stomach ulcers. The study by Elliott et al. [124] gave rise to the concept of probiotic use. Gram-negative bacteria quickly colonized the ulcer site in a rat model of acetic acid-induced stomach ulcer, which seriously hampered ulcer healing. Gram-positive bacterial colonization, however, aided in the healing of ulcers. Notably, ulcer healing was hastened by administering the exogenous probiotic strain Lactobacillus.

Table (8). Effect of dietary intervention with pomegranate and alovera fortified with yogurt on %Ulceration, Preventive Index and Severity Ulcer of induced peptic ulcer rats.

	Ulceration(%)		PI (%)		SOU	
Groups	Mean±SD	Sig	Mean±SD	Sig	Mean±SD	Sig
NGG	0.000e±0.00		--		--	
PGG	82.90a±4.70		15.92d±1.81		5.57a±0.49	
AVG	48.80c±2.00	0.000	47.93b±3.57	0.000	2.76b±1.30	0.000
PMG	60.26b±3.68		37.16c±7.29		3.43b±0.53	
APG	35.87d±5.89		60.17a±5.24		1.31c±0.62	
LSD	10.467		8.717		0.917	

*Values are expressed as mean±SD, means in the same columns with different letter are significantly (P≤0.05), LSD: Least significant of difference, *NS: Non significant; PI: Preventive Index; SOU: Severity of Ulcer ; NGG: negative control (-ve) group; PGG: positive control (+ve) group; AVG: alovera fortified with yogurt; PMG: Pomegranate fortified with yogurt ; APG: alovera and pomegranate fortified with yogurt.*

There was sufficient evidence to suggest that probiotic consumption can help patients with stomach ulcers, lowering inflammation, and accelerating gut healing, even though the evidence supporting the direct role of yogurt and probiotics in ulcer treatment is still developing. Probiotics and yogurt should be used as part of a comprehensive treatment strategy that may also include proton pump inhibitors or antibiotics.

Histopathological examination of stomach:

Microscopically, stomach of rats from group 1 revealed the normal histological architecture

of gastric layers (Figs. 1 & 2). In contrast, stomach of rats from group 2 showed remarkable histopathological alterations characterized by focal necrosis of gastric mucosa, mucosal inflammatory cells infiltration, submucosal edema associated with inflammatory cells infiltration (Figs. 3, 4, 5 & 6). Meanwhile, ameliorative effect was noticed in the stomach of rats from group 3; some examined sections from groups 3 exhibited slight submucosal edema (Figs. 7 & 8), whereas other sections from this group showed inflammatory cells infiltration in the mucosa (Fig. 9) and submucosa associated with submucosal edema (Figs. 9 & 10). Furthermore, some stomach sections of rats from group 4 exhibited no histopathological lesions (Figs. 11& 12), whereas other sections revealed inflammatory cells infiltration in the mucosa (Fig. 13), submucosal edema (Figs. 13 & 14) and submucosal few inflammatory cells infiltration (Fig. 14). Moreover, most examined gastric tissues of rats from group 5 described no histopathological lesions (Figs. 15, 16 & 17), whereas few sections showed focal necrosis of gastric mucosa (Figs. 18 & 19) associated with inflammatory cells infiltration (Fig. 19)

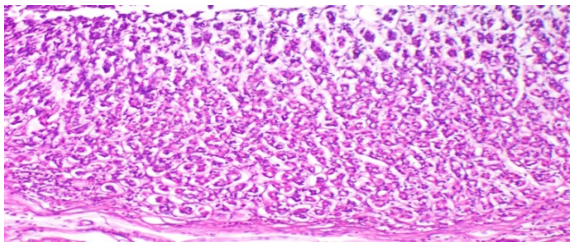


Fig. (1): Photomicrograph of stomach of rat from group 1 showing the normal histological architecture of gastric layers (H & E stain, X 100).

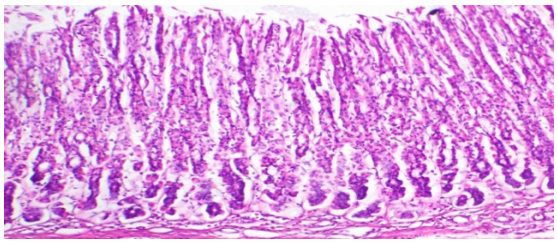


Fig. (2): Photomicrograph of stomach of rat from group 1 showing the normal histological architecture of gastric layers (H & E stain, X 100).

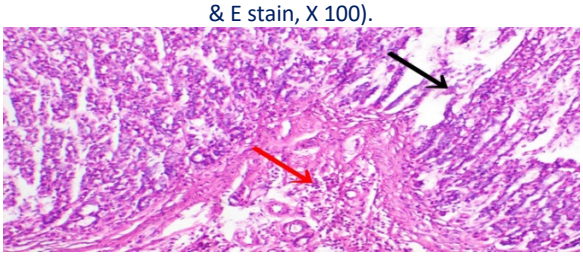


Fig. (3): Photomicrograph of stomach of rat from group 2 showing focal necrosis and sloughing of gastric mucosa (black arrow) associated with submucosal inflammatory cells infiltration (red arrow) (H & E stain, X 100).

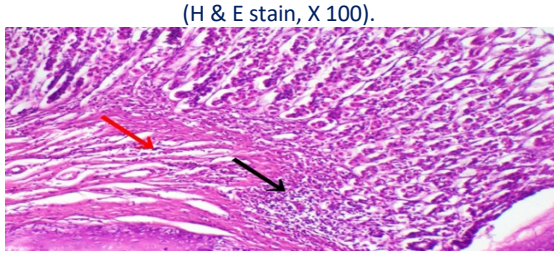


Fig. (4): Photomicrograph of stomach of rat from group 2 showing inflammatory cells infiltration in the mucosa (black arrow) and submucosa (red arrow) (H & E stain, X 100).

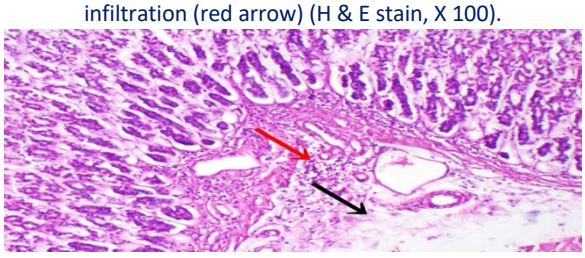


Fig. (5): Photomicrograph of stomach of rat from group 2 showing submucosal edema (black arrow) associated with inflammatory cells infiltration (red arrow) (H & E stain, X 100).

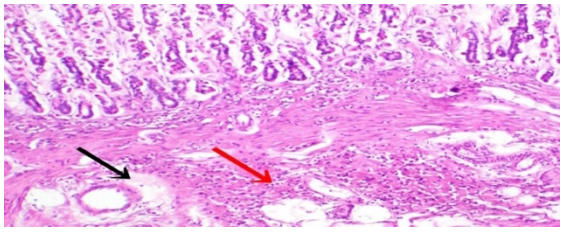
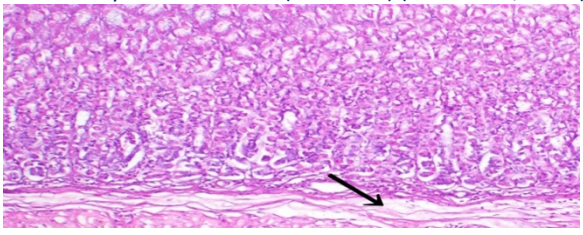


Fig. (6): Photomicrograph of stomach of rat from group 2 showing submucosal edema (black arrow) associated with inflammatory cells infiltration (red arrow) (H & E stain, X 100).



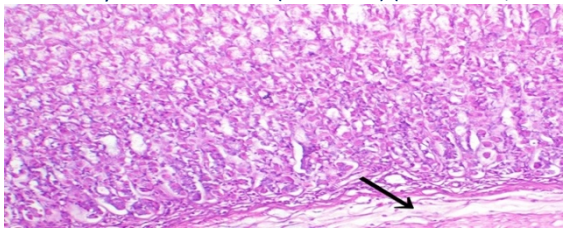


Fig. (7): Photomicrograph of stomach of rat from group 3 showing slight submucosal edema (arrow) (H & E stain, X 100).

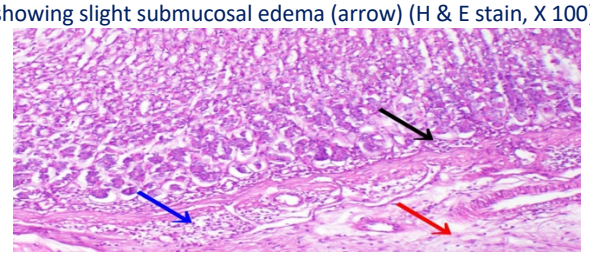


Fig. (8): Photomicrograph of stomach of rat from group 3 showing slight submucosal edema (arrow) (H & E stain, X 100).

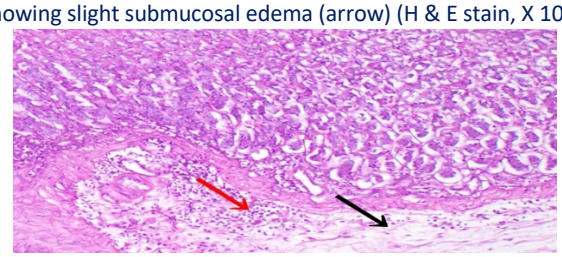


Fig. (9): Photomicrograph of stomach of rat from group 3 showing inflammatory cells infiltration in the mucosa (black arrow) and submucosa (red arrow) associated with submucosal edema (blue arrow) (H & E stain, X 100).

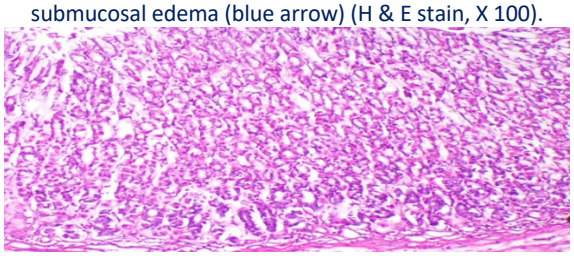


Fig. (10): Photomicrograph of stomach of rat from group 3 showing submucosal edema (black arrow) associated with inflammatory cells infiltration (red arrow) (H & E stain, X 100).

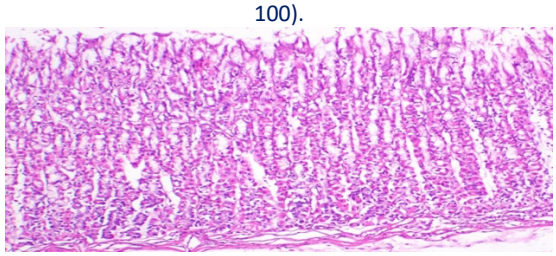


Fig. (11): Photomicrograph of stomach of rat from group 4 showing no histopathological lesions (H & E stain, X 100).

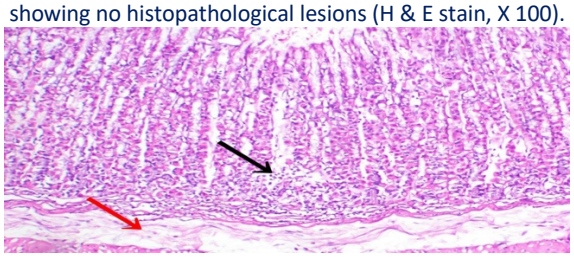


Fig. (12): Photomicrograph of stomach of rat from group 4 showing no histopathological lesions (H & E stain, X 100).

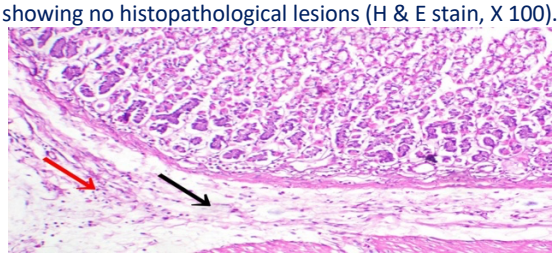


Fig. (13): Photomicrograph of stomach of rat from group 4 showing inflammatory cells infiltration in the mucosa (black arrow) associated with submucosal edema (red arrow) (H & E stain, X 100).

Fig. (14): Photomicrograph of stomach of rat from group 4 showing with submucosal edema (black arrow) associated with inflammatory cells infiltration (red arrow) (H & E stain, X 100).

Fig. (15): Photomicrograph of stomach of rat from group 5 showing no histopathological lesions (H & E stain, X 100).

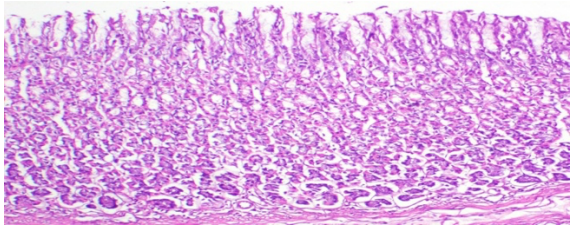


Fig. (16): Photomicrograph of stomach of rat from group 5 showing no histopathological lesions (H & E stain, X 100).

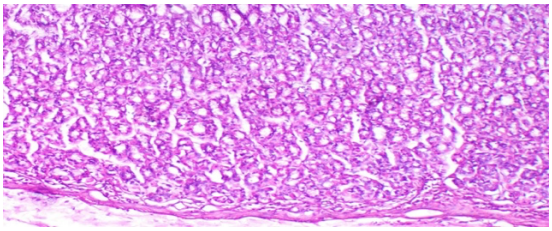


Fig. (17): Photomicrograph of stomach of rat from group 5 showing no histopathological lesions (H & E stain, X 100).

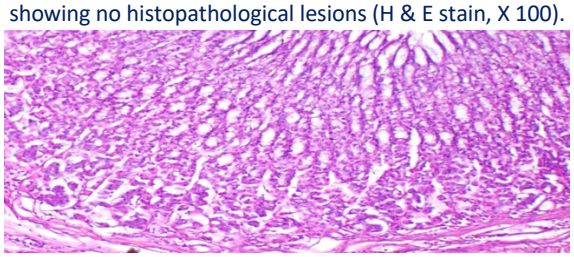
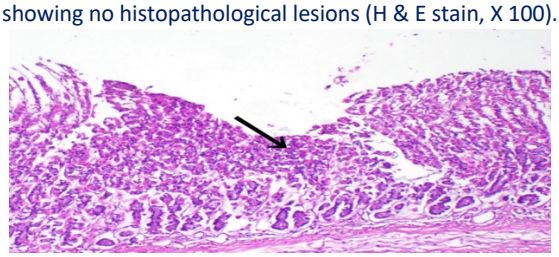


Fig. (18): Photomicrograph of stomach of rat from group 5 showing focal necrosis of gastric mucosa (black arrow) (H & E stain, X 100).



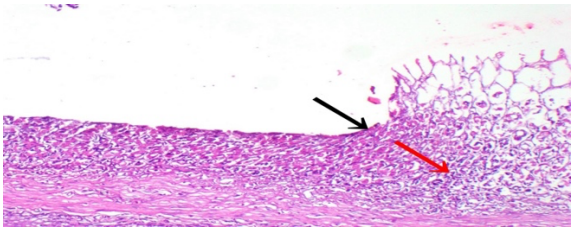


Fig. (19): Photomicrograph of stomach of rat from group 5 showing focal necrosis of gastric mucosa (black arrow) associated with inflammatory cells infiltration (red arrow) (H & E stain, X 100).

4. Conclusion

The study demonstrates that dietary intervention with aloe vera, pomegranate, and their combination significantly improved hematological parameters, oxidative stress markers, and ulcer healing in peptic ulcer-induced rats. The combined treatment group (APG) exhibited the most notable enhancements, including increased hemoglobin levels, RBC count, and antioxidant enzyme activity (SOD and CAT), alongside a significant reduction in malondialdehyde (MDA) levels. Additionally, combination formula achieved the highest ulcer protection rate and the most effective inhibition of the ulcer index, suggesting a synergistic effect of aloe vera and pomegranate in promoting gastric mucosal healing. These findings indicate that the combined dietary supplementation could be a promising natural therapeutic strategy for managing peptic ulcers.

Conflict of Interest

The authors declare that there is no conflict of interest.

Funding

No fund has been received.

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الترقيم الدولي اون لاين الترقيم الدولي للطباعة
2735-5934 2735-590X

Received: 30 Sep 2024
Accepted: 22 Oct 2024

تاريخ الاستلام: ٥ فبراير ٢٠٢٥
تاريخ القبول: ١٦ مارس ٢٠٢٥
تاريخ النشر: ١ ابريل ٢٠٢٥

نوع المقالة: بحوث اصلية
التغذية وعلوم الاطعمة

تأثير التدخل الغذائي بالرمان والصبار في تخفيف اعراض قرحة المعدة المستحثة بالكحول في فئران التجارب

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الملخص العربى:
يتميز مرض القرحة الهضمية بتقرحات و التهابات فى المعدة والاثنى عشر، وغالبًا ما ترتبط بالإجهاد التأكسدي والالتهاب. هدفت هذه الدراسة إلى تقييم التأثير العلاجى لخليط معزز بمستخلص قشر الرمان و جيل الصبار فى نموذج قرحة مستحث تجريبياً فى الفئران البينو. تم تقسيم خمسة وثلاثين فأرًا ذكرًا بالغًا إلى مجموعة تحكم سلبية ومجموعات مستحثة بالقرحة، تلقت جرعة فموية واحدة من الكحول الإيثيلى بنسبة 10 مل / كجم من وزن الجسم، وتم تقسيمها عشوائيًا إلى أربع مجموعات فرعية؛ والتى تلقت علاجات مختلفة: 200 مجم / كجم من الصبار (AVG)، و 50 مجم / كجم من الرمان (PMG)، ومجموعة مشتركة من الصبار والرمان (APG). أظهرت النتائج أن التدخلات الغذائية حسنت بشكل كبير من مؤشرات صورة الدم ومضادات الأكسدة. أظهرت مجموعة APG أعلى مستويات الهيموجلوبين (12.25 ± 0.70 جم / ديسيلتر) وعدد خلايا الدم الحمراء (7.03 ± 0.59)، مما يدل على تحسن كبير مقارنة بمجموعة التحكم الموجبة. كما تم تعزيز نشاط مضادات الأكسدة، مع أعلى مستوى من سوبر اكسيد الدسميوتاز (SOD) (25.03 ± 0.55 وحدة / مل) وانخفاض مستويات مالوندیالدهيد (MDA) (1.54 ± 0.53 نانومول / مل) فى APG. كان مؤشر تثبيط القرحة هو الأعلى فى APG (359.71 ± 44.14)، إلى جانب أعلى معدل حماية (60.17 ± 5.24٪) وأقل نسبة تقرح (35.87 ± 5.89٪). تسلط هذه النتائج الضوء على التأثيرات الوقائية لصبار الألوفيرا ومستخلص قشر الرمان فى تقليل تقرح المعدة وتعزيز دفاع مضادات الأكسدة وتعزيز التئام القرحة. وأظهر العلاج الخليط الفوائد العلاجية الأكثر تأثيرا، مما يشير إلى إمكانية استخدامه كاستراتيجية طبيعية للتخفيف من قرحة المعدة وتحسين صحة الجهاز الهضمي.

الكلمات الكاشفة: المعدة، البريبايوتيك، مضادات الأكسدة، الالتهاب، مؤشر الحماية، درجة التقرح

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Gharib et al., 2025, The effect of dietary intervention with pomegranate and aloe vera in alleviating the symptoms of alcohol-induced stomach ulcers in experimental rats. JHE, 35 (2), 57-84. DOI: 10.21608/mkas.2025.365849.1373