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Original Article

The protective role of camel's milk on some hematological parameters of male rats infected with gastric ulcer

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ABSTRACT

The present study was carried out to investigate the protective role of camel's milk in some hematological parameters of male rats treated with ethanol (80%) to induce acute gastric lesions. Male rats weighted (280 – 300 g)were divided in five groups : (control group (I)) that were treated with (5ml/kg) of physiological saline (0.85% NaCl), (milk group (II)) that were treated with (5ml/kg) of raw camel's milk, (ulcer group (III)) that were treated with (5ml/kg) of ethanol (80%) and sacrificed after(24 hours), (ulcer group(IV)) that were treated with (5ml/kg) of ethanol (80%) and after (24 hours) treated with (5ml/kg) of physiological saline,(treatment group (V)) initially treated with (5ml/kg) of ethanol (80%) then after (24 hours) treated with (5ml/kg) of raw camel's milk. The results showed a significant decrease($P \leq 0.05$) in (RBC , Hb , PCV , MCH and MCHC) in the ulcer groups(III) and (IV),while there was showed a significant increase ($P \leq 0.05$)in (WBC, MCV) compared with control group(I), milk group (II) and treatment group (V).

Key word: Ethanol, Gastric lesions, Camel' s milk, Hematological parameters.

INTRODUCTION

Gastric ulcer is a chronic disease that affects millions of people worldwide which involves disruption in the skin or mucus membrane lining alimentary canal [1].The basic pathophysiological of gastric ulcer results from an imbalance between some endogenous aggressive factors [hydrochloric acid, pepsin, refluxed bile, leukotrienes, reactive oxygen species (ROS)] and cytoprotective factors, which include the function of the mucus-bicarbonate barrier, surface active phospholipids, prostaglandins (PGs), mucosal blood flow, cell renewal and migration, non-enzymatic and enzymatic antioxidants, and some growth factors [2,3]. Alcohol, smoking, nutritional deficiencies and frequent ingestion of nonsteroidal anti-inflammatory drugs (NSAIDs) factors are contribute to induce gastric ulcer [4]. Spicy food, coffee and emotional stress factors that able to increase the acid secretion of stomach and causing the pain of an existing ulcer to be worse, smoking don't cause gastric ulcer but it increase chance of getting an ulcer and slow the healing of existing ulcer, in rare case stomach tumors can causes ulcers [5,6]. Gastric ulcers are also associated with considerable morbidity related to chronic epigastric pain, nausea, vomiting, and anemia [7]. Intake of ethanol induces the overproduction of reactive oxygen species (ROS) and the decrease in the activity of antioxidant enzymes, such as catalase (CAT), glutathione S-transferase (GST), glutathione peroxidase (GPx), superoxide dismutase (SOD), and glutathione reductase (GR), leading to gastric mucosal injuries, including ulceration, erosion, hemorrhage, congestion, and edema [8,9]. Camel's milk (CM) is different from other ruminant milk, it is low in cholesterol, sugar and protein but high in minerals (sodium, potassium, iron, copper, zinc and magnesium), vitamins (A, B2, C and E), and contains a high concentration of insulin [10]. The (CM) is an excellent source of well-balanced nutrients and also exhibits a range of biological activities that influence digestion, metabolic responses to absorbed nutrients, growth and development of specific organs and resistance to diseases. These biological activities are mainly due to the presence of peptides and protein in milk [11,12]. The milk is considered to have medicinal properties. The products developed also include cosmetics or pharmaceuticals. A series of metabolic and autoimmune diseases are successfully being treated with camel's milk. In India, camel's milk is used therapeutically against dropsy, jaundice, problems of the spleen, tuberculosis, asthma, anemia, piles

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and diabetes. Antibacterial and antiviral activities of these proteins of camel's milk were studied [13,14]. The Present study aimed to investigate the protective role of camel's milk on some hematological parameters of male rats treated with ethanol to induce acute gastric ulcer.

MATERIALS AND METHODS

Camel's Milk: Camel's milk samples were collected in the morning from a herd of camels, from different parts of the city of Nasiriyah / Thi_Qar province, Iraq which lies (360 km) from the province of Baghdad by using hand milking in sterile screw bottles and then transferred directly mediated icebox refrigerated to the laboratory and keeping it in the refrigerator under temperature (5-2) º C until use.

Animals: Thirty adult healthy male rats (Rattusnorvegicus) weighing (280-300g) were used in the present study. Animals were housed in the animal house of Biology Dept., Science College, Thi-Qar University, Iraq. Animals were housed in plastic boxes with aluminum cover bedded with wooden chips. The animals were housed under standard laboratory conditions (12h light: 12h dark photoperiod cycle (LD) at (20 ± 2) º C and relative humidity (45-55) %). Animals were fed on standard rat pellet (ad *libitum*) and tap water, the animals left for two weeks for the purpose of acclimatization before the start of the experiment.

Experimental group's protocol: The male rats divided into five groups, each group containing (6) animals, upon the following:

Group I	Control (normal) that were treated with (5ml/kg) of physiological saline (0.85% Nacl), for three weeks.
Group II	Milk group (normal) that were treated with (5ml/1Kg) of raw camel's milk, for three weeks.
Group III	(Ulcer group 1) that were treated with (5ml/kg) of ethanol (80%) for induction of ulcer and sacrificed after (24 hours) from treated. According to the method described by [15] with slight modification.
Group IV	(Ulcer group 2) that were treated with (5ml/kg) of ethanol (80%) for induction of ulcer and after (24 hours) treated with (5ml/kg) of physiological saline daily for three weeks.
Group V	(Treatment group) initially treated with (5ml/kg) of ethanol (80%) for induction of ulcer then after (24 hours) of treatment with ethanol it treated with (5ml/kg) of raw camel's milk daily for three weeks.

Induction of gastric ulcer by ethanol: The animals were weighted before starting the experimental and it starved for (48) hours before use to ensure an empty stomach with free access water [16].During the fasting period the rats were placed individually in separate cage. After (48) hours the animals in groups (III, IV, V) treated with (5ml/kg) ethanol (80%) orally by gastric gavage needles, [15] with slight modification (the modification include the dose of ethanol used (5ml/kg) instead of (10ml/kg).

Blood collection: After the end of the experimental period the animals euthanized by ether (BDH, England) and sacrificed by abdominal opened and collected blood samples for each animal directly from the heart in a way cardiac puncture.Blood samples drawn from the animals using syringes medical capacity (5ml) after the withdrawal of blood samples put part of it (1ml) in the tube contain anti-clotting (EDTA) to measuring hematologicalparameters.

Hematological parameters: Hematological parameters measuring by using hematology analyzer (Nihon Kohden)corporation (Japan) in the laboratories of the blood bank in the city of Nassiriyah, Iraq which included:red blood cells (RBC), hemoglobin (Hb), packed cell volume (PCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), mean corpuscular volume(MCV) and white blood cells (WBC).

Statistical analysis: ANOVA analysis and LSD tests were used according to (SPSS Version 16) program at the ($p<0.05$)to find the mean between all treatments.

RESULTS AND DISCUSSION

The results showed non-significant differences($P \leq 0.05$)in some hematological parameters (RBC, Hb, PCV, MCH, MCHC, MCV, WBC) of the milk group (II) compared with control group (I).The ulcer groups (III) and (IV) showed a significant decrease ($P \leq 0.05$) in some hematological parameter, (RBC, Hb, PCV, MCH and MCHC),compared with control group(I), milk group(II) and treatment group (V).This decrease may be caused by Increase (RBC) hemolysis after acute or chronic ethanol administration has been

ascribed to many factors. These include enhanced osmotic hemolysis due to increase erythrocyte fragility and impairment of the antioxidant defense mechanisms of the cell membrane, especially by changes in the lipid fluidity of the middle zone of the bilayer [17].Thus, the reduction of (RBC and Hb) count in ethanol treated rats might be a reflection of oxidative stress effects of ethanol on red cell membrane. Or may be due to bone marrow suppressive effect of ethanol [18]. Ethanol have suppressive effect on bone marrow cells,and suppresses marrow granulopoiesis. It was may be causes decrease in the level of the packed cell volume (PCV) in the blood, and particularly in the generation of (RBC), causing the low number of red blood cells which is reflected in the average (PCV) [18,19]. The treatment group (V) showed non-significant differences in (RBC, HGB, PCV, MCH and MCHC) compared with control group (I)($P \leq 0.05$) and a significant increase ($P \leq 0.05$) in this parameters compared with ulcer groups (III) and (IV).The decrease in the (RBC) hemolysis in the (V) group related with presence of vitamin (E) in camel's milk and is this vitamin is an essential part of cell membranes, a site to defend the cell against oxidation and plays a major role in maintaining the flexibility of red blood cells and reduce the fragility and damage as a result of the oxidation of phospholipid membranes red blood cells [20],this vitamin also has a direct impact on the process of formation of red blood cells in the bone marrow referred [21],to the effect of vitamin (E) in the metabolism of vitamin (B_{12}), which is necessary for the bone marrow to produce blood cells intact.It also raise the rate of the concentration of hemoglobin by increasing the activity of enzymes chain formation of hemoglobin, including the enzyme (ALAD) as well as raise the readiness of some of the basic elements in the manufacture of hemoglobin such as iron and copper [22],Also the treated with raw camel's milk caused rises the packed cell volume (PCV) compared with the groups that treated with ethanol this result reflects the significant improvement that showed by this group at the level of the total number of (RBC) and (Hb) that led to improve the blood indicators (MCH,MCHC and MCV) because it was effected with the value of (RBC, Hb and PCV)[23]. On the other hand the ulcer groups (III) and (IV) showed a significant increase ($P \leq 0.05$)in(MCV) and (WBC) compared with control group (I), milk group(II) and treatment group(V).The increase in (MCV) may be caused by the anemia was macrocytic and hypochromic as shown by decrease total (RBC) count, decreased (Hb) and (MCHC) concentrations, increased (MCV) [24]. And the total (WBC) increased may be according to the role of (WBC)as a defense of the body against inflammatory processes occurring in the lining of the digestive tract. The treatment group (V) showed a significant decrease **($P \leq 0.05$)** in (MCV)and total (WBC) compared with ulcer groups (III) and (IV).This decrease in (MCV) was may bethe treated with raw camel's milk caused rises the packed cell volume (PCV) compared with the groups that treated with ethanol this result reflects the significant improvement that showed by this group at the level of the total number of (RBC) and (Hb) that led to improve the blood indicators (MCH,MCHC,MCV) because it was effected with the value of (RBC, Hb, PCV)[25].The decrease intotal (WBC) may be caused by theprotective effect of camel's milk against ethanol induced oxidative stress was due it have antioxidant properties, camel's milk was found to contain high concentrations of vitamins (A,B2,C and E) and is very rich in magnesium and other trace elements, these vitamins act as antioxidants and have been found to be useful in preventing toxicant-induced tissue injury [25].which may be lead to reduce the inflammatory tissues and decrease the total (WBC).

Table (1): The role of camel's milk on hematological parameters of male rats infected with gastric ulcer.

GROUPS	RBC(10^{12}/L)	HGB(g/dl)	PCV(%)	MCH(p g)	MCHC(g/dl)	MCV(μm^3)	WBC(10^9/L)
GROUP I	8.16^{ab} ±0.05	14.31^{a} ±0.14	45.11^{ab} ±0.07	18.95^{a} ±0.09	31.66^{a} ±0.34	56.66^{b} ±0.50	8.21^{c} ±0.08
GROUP II	8.35^{a} ±0.01	15.19^{a} ±0.35	46.49^{a} ±0.29	19.13^{a} ±0.12	32.96^{a} ±0.17	56.68^{b} ±0.27	8.53^{c} ±0.20
GROUP III	6.14^{c} ±0.01	11.93^{b} ±0.09	40.45^{c} ±0.62	16.48^{b} ±0.11	27.85^{c} ±0.38	60.33^{a} ±0.47	18.08^{a} ±0.31
GROUP IV	5.12^{d} ±0.00	11.21^{b} ±0.13	39.01^{d} ±0.11	16.11^{b} ±0.01	27.08^{c} ±0.62	61.11^{a} ±0.25	14.01^{b}±0.15
GROUP V	8.03^{b} ±0.06	14.08^{a} ±0.07	44.51^{b} ±0.20	18.56^{a} ±0.13	32.03^{a} ±0.17	57.03^{b} ±0.47	8.61^{c} ±0.05
LSD	**0.31**	**1.50**	**1.40**	**0.83**	**1.27**	**1.04**	**4.03**

Values are means ± S.E; Different letters refer to significant differences ($p \leq 0.05$); same letters refer to non significant differences ($p \leq 0.05$); RBC = Red Blood Cells, HGB = Hemoglobin, PCV= Packed Cell Volume, MCH= mean corpuscular hemoglobin, MCHC= mean corpuscular hemoglobin concentration, MCV= mean corpuscular volume, WBC = White Blood Cells

CONCLUSION

Our results demonstrate that ethanol is capable of inducing marked alterations in some hematological parameters and oxidative damage, and inhibiting the function of antioxidant enzymes. Drinking camel's milk could be beneficial for alleviating ethanol toxicity.

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