APPLIED MEDICAL RESEARCH, 2018
VOL 4, NO. 1, PAGE 12–17
10.5455/amr.20180724045336

eJManager

ORIGINAL ARTICLE

Open Access

Effect of camel milk supplementation in management of gastric ulcer

Aminu Lailaba Abubakar[1], Abdullahi Dandare[1], Shamsudeen Umar Dandare[1], Sulaiman Rabiu[1], Abdulqadir Sule Ibrahim[2], Saifullahi Armaya'u[1]

[1]Department of Biochemistry, Usmanu Danfodiyo University, Sokoto, Nigeria

[2]Department of Ophthalmology, Usmanu Danfodiyo Teaching Hospital, Sokoto, Nigeria

ABSTRACT

Background: Gastric ulcer is one of the major causes of morbidity globally. It is prevalent in Nigeria and more frequent among the lowest income group; this is primarily linked to their standard of living and constant use of non-steroidal anti-inflammatory drugs. The use of camel milk in the treatment of many diseases has long been established.

Objective: This study was designed to evaluate the potentials of camel milk in the treatment of gastric ulceration.

Methods: Albino rats weighed 120–150 g were divided into four groups of five rats each. Group I: negative control, group II: positive control, group III: ethanol-induced gastric ulcer rats supplemented with camel milk, group IV: ethanol-induced gastric ulcer rats orally dosed with omeprazole.

Results: The results of this study revealed that white blood cells, red blood cells, HBC, pack cell volume, mean corpuscular volume, and platelets decreased significantly ($P < 0.05$) in ethanol-induced ulcer group compared to other groups. There were no significant differences ($P > 0.05$) in mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration level for all the groups investigated. The levels of antioxidant enzymes (catalase and superoxide dismutase) and vitamins (A, C, and E) were significantly improved toward normal ($P < 0.05$) due to camel milk supplementation. The histopathological analysis of the stomach tissue indicates preserved transmural architecture with clearly defined mucosa, submucosa, and muscularis propria in camel milk supplemented group and omeprazole treated group.

Conclusion: These results demonstrated the potential use of camel milk in reversing the damaging effect of ethanol-induced gastric ulcer and alteration of some biochemical parameters. Hence, camel milk could be used as a dietary supplement for managing gastric ulcer.

ARTICLE HISTORY

Received 24 July 2018
Accepted 29 August 2018
Published 05 September 2018

KEYWORDS

Camel milk; ethanol; gastric ulcer; omeprazole; transmural architecture

Introduction

Ulcer is a life-threatening disease that affects millions of people globally. It is characterized by a disruption of mucus membrane lining of alimentary canal [1]. The basic pathophysiology of gastric ulcers results from an imbalance between some endogenous factors such as hydrochloric acid, pepsin, refluxed bile, leukotriene, reactive oxygen species (ROS) etc., and cellular protective factors such as mucus bicarbonate barrier, phospholipids, mucosal blood flow, cell renewal and migration, and antioxidants [2]. Alcoholism, smoking, nutritional deficiencies, and frequent ingestion of non-steroidal anti-inflammatory drugs contribute to gastric ulcers [3]. Spicy food, coffee, and emotional stress are factors that are able to increase the acid secretion of the stomach and causing the pain of an existing ulcer [4].

Despite the availability of antiulcer drugs, there are increasing cases of ulcer in Nigeria probably as a result of the economic situation of the nation or due to the limited access to drugs, particularly to the rural dwellers. In addition, most of the common antiulcer drugs possess some side effects.

Contact Abdullahi Dandare youngdandare@gmail.com Department of Biochemistry, Usmanu Danfodiyo University, Sokoto, Nigeria.

Therefore, it is imperative to continue searching for safe, available, and affordable means of ulcer treatment.

Camel milk is available and accessible in most of the Nigerian rural communities, particularly in the northern part of the country. The milk is highly nutritious and has been used to cure various forms of diseases [5]. Therefore, this work was designed to investigate the therapeutic potentials of the camel milk against gastric ulcer.

Materials and Methods

Camel milk collection

Camel milk was obtained early in the morning from lactating camel at Kasuwar Daji kara area Sokoto, Nigeria. It was collected in sterile containers, transported to the laboratory, and kept in a refrigerator (4°C) till further use.

Experimental animals

Twenty adult rats (*Rattus norvegicus)* weighing (120–150 g) were used in the present study. The animals were housed in plastic cages with aluminium cover bedded with saw dust and kept under standard laboratory conditions. The rats were fed on growers vital feed and supplied with constant tap water, then allowed to acclimatize for 2 weeks prior to the commencement of the research.

Experimental group protocol

The animals were divided into four groups of five rats each and treated as follows:

Group 1 Negative control: placed on distilled water

Group 2 Positive control: Ethanol-induced ulcer untreated

Group 3 Ethanol-induced ulcer treated with 5 ml/kg of camel milk [6]

Group 4 Ethanol-induced ulcer treated with 20 mg/kg omeprazole: a product of Navketan Pharma Pvt. Ltd./India) [7]

Ulcer Induction

The rats in groups (I–IV) were starved for 48 hours with free access to water to ensure an empty stomach, then the animals in groups II, III, and IV were orally dosed with 80% ethanol (5 ml/kg body weight) through the gastric gavage needles and allowed for 24 hours for the onset of ulcers [8]. Then, the camel milk and omeprazole treatments were followed accordingly for 28 days.

Sample Preparation

Twenty-four (24) hours after the last treatment, the animals were anesthetized with chloroform vapor, and then sacrificed under the supervision of a veterinarian. The blood samples were collected and prepared for biochemical investigations. The stomach was harvested, washed with normal saline, and stored in 10% formalin for histopathological studies.

Histopathological Evaluation

The harvested stomach was fixed in formalin for 24 hours. The tissue was then fixed in 10% buffered formalin and processed using a tissue processor. The slides were examined for morphological changes [9].

Hematological Analysis

Samples were analyzed with an hematology auto analyzer (PCE-210 N; Erma, Inc, Tokyo, Japan), Analyses were performed according to the standard operating manual.

Determination of Antioxidant Enzymes

Catalase (CAT): Determination of CAT activity was achieved by the method described by Beers and Sizer [10].

Superoxide dismutase (SOD): The activity of SOD was measured according to the method described by Zhou et al. [11].

Vitamins A, C, and E concentrations were assayed according to the method described by Rutkowski and Grzegorczyk [12].

Data Analysis

Data generated were presented in tabular form and expressed as mean ± standard deviation.

The values were tabulated and presented as mean ± standard error of the mean. The results were statistically analysed by one-way ANOVA using Graph Pad Instat Software, Version 3.0, San Diego, CA, USA. Tukey multiple comparison was used to compare the difference between the means. The differences were considered statistically significant at $P < 0.05$.

The results of the histopathological studies were presented in the form of photomicrographs.

Table 1. Effect of camel milk supplementation on hematological parameters in ethanol-induced ulcer rats.

Parameters	Group I	Group II	Group III	Group IV
WBC ($\times10^3$l)	6.13 ± 0.73[a]	15.88 ± 0.73[b]	11.03 ±1.27[c]	10.77 ± 2.83[c]
RBC ($\times10^6$l)	5.63 ± 0.35[a]	3.85 ± 0.17[b]	6.41 ± 0.26[a]	5.01 ± 0.36[a]
HGB (g/l)	12.33 ± 0.33[a]	5.47 ± 0.79[b]	13.17 ± 0.84[a]	10.23 ± 0.88[a]
PCV (%)	36.27 ± 2.05[a]	16.37 ± 2.19[b]	36.60 ± 1.17[a]	29.91 ± 1.59[c]
MCV (fl)	65.67 ± 1.68[a]	58.20 ± 1.09[b]	57.17 ± 1.15[b]	60.00 ± 1.56[ab]
MCH (pg)	21.97 ± 1.39[a]	19.10 ± 0.59[a]	20.57 ± 1.27[a]	20.50 ± 1.19[a]
MCHC (g/dl)	33.33 ± 1.47[a]	28.50 ± 5.26[a]	35.93 ± 1.69[a]	34.10 ± 1.59[a]
PLT ($\times10^3$l)	250.33 ± 55.48[a]	406.67 ± 77.74[b]	604.67 ± 55.44[c]	309.33 ± 82.18[d]

WBC- White blood cells, RBC- Red blood cells, HGB- Haemoglobin, HCT- Haematocrit, MCV- Mean corpuscular volume, MCH- Mean corpuscular haemoglobin, MCHC- Mean corpuscular haemoglobin concentration, PLT- Platelets. Values with different superscript (a, b, c, d) in rows are statistically significant (p<0.05) while values with the same superscript are statistically non significant (P>0.05).

Table 2. Effect of camel milk supplementation on antioxidant vitamins and enzymes in ethanol-induced ulcer rats.

Groups	Vitamin A (µmol/l)	Vitamin C (µmol/l)	Vitamin E (mg/dl)	CAT (units/ml)	SOD (units/ml)
I	1.77 ± 0.16[a]	19.79 ±3.94[a]	819.08 ± 188.31[a]	28.71 ± 4.52[a]	135.93 ± 14.42[a]
II	1.22 ± 0.49[a]	27.47 ± 5.22[b]	615.35 ± 210.53[b]	27.63 ± 3.54[a]	104.21 ± 7.77[b]
III	1.21 ± 0.09[a]	34.69 ± 4.82[cb]	797.55 ± 20.33[c]	44.93 ± 5.53[b]	306.53 ± 62.84[c]
IV	1.45 ± 0.31[a]	36.25 ± 0.42[c]	778.72 ± 49.72[d]	51.07 ± 3.22[b]	306.52 ± 52.85[c]

SOD: superoxide dismutase, CAT: catalase. Values with different superscript in a row are statistically significant (p < 0.05) while values with the same superscript are statistically non-significant (P > 0.05).

Results

Table 1 shows the results of hematological parameters of ethanol-induced ulcer rat model treated with camel milk. It was observed that the level of white blood cells (WBC), red blood cells (RBC), hemoglobin count (HBC), pack cell volume (PCV), mean corpuscular volume (MCV), and platelets (PLT) decreased significantly (P < 0.05) in ethanol-induced ulcer group compared to other groups. However, no significant difference (P < 0.05) was observed in levels of MCV in the ulcer-induced untreated group compared to the camel milk and omeprazole treated group. The levels of all hematological parameters were investigated with the exception of WBC, MCV, and PLT that remained similar in camel milk treated group compared to the control (see Table 1). It was also observed that the mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were similar among all the groups (P > 0.05)

Table 2 shows the results of antioxidant enzymes and vitamins of ethanol-induced ulcer rat model treated with camel milk. It was noted that the levels of vitamin A remain statistically similar (P < 0.05) among the groups analyzed. The concentration of vitamin C increased significantly (P < 0.05) due to omeprazole treatment and camel milk supplementation while the concentration of vitamin E is significantly decreased (P > 0.05) due to ulcer induction. However, the administration of camel milk and omeprazole improves its concentration towards normal. CAT and SOD activities increased significantly in omeprazole and camel milk treated groups compared to positive and negative control groups. It was also observed that the increase in CAT activity is similar in omeprazole treated group compared to camel milk treated group (see Table 2).

Figures 1–4 are micrographs of the stomach sections of the normal control group, ulcer-induced untreated group, ulcer-induced treated with camel milk, and ulcer-induced treated with omeprazole, respectively. Ulcer-induced untreated group (Figure 2) shows mild ulceration evidence from a discontinuity in the mucosa while other groups (see Figures 1, 3, and 4) show no stomach lesion evidence from a preserved transmural architecture with clearly defined mucosa, submucosa, and muscularis propria.

Discussion

This study was conducted to investigate the potential of camel milk supplementation in the management of gastric ulcer. Increase production of ROS and/or a decrease in antioxidant levels causes oxidative stress, which plays an important role in the pathogenesis of gastric ulcer [13]. The results of this study revealed that ingestion of 80% ethanol not only causes a lesion in the gastric tissue of the experimental rats but also drastically decreases the level of antioxidant vitamins and enzymes. Interestingly,

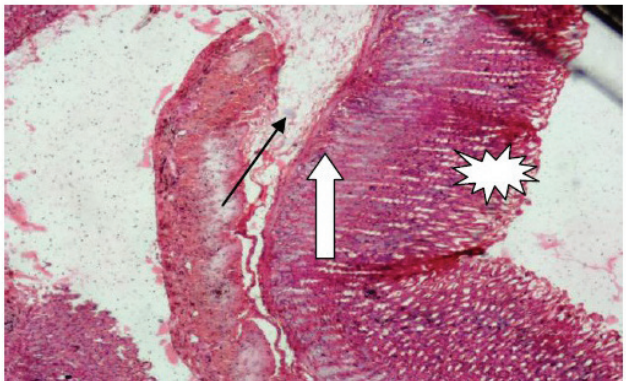

Figure 1. Negative control: section of the stomach howing mucosa (white asterisk), submucosa (white arrow), and muscularis propria (black arrow). Hematoxylin & Eosin (H & E) × 40 magnification.

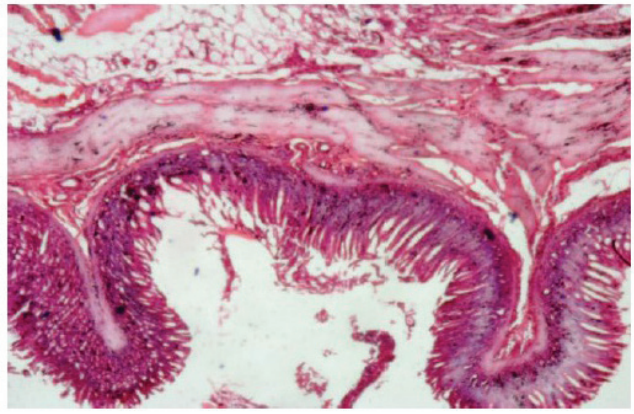

Figure 3. Camel milk treated group: section of the stomach showing preserved Transmural Architecture. H & E × 40 magnification.

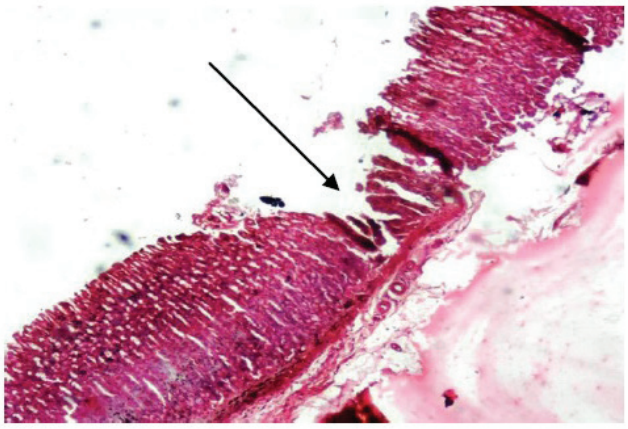

Figure 2. Positive control. Section of the stomach showing a discontinuity in the mucosa (mild-moderate ulceration) H & E × 40 magnification.

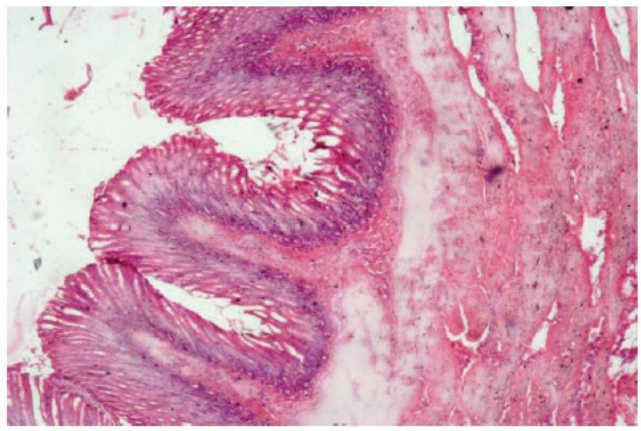

Figure 4. Omeprazole treated control. Section of the stomach showing preserved transmural Architecture. H & E × 40 magnification.

camel milk administration had revert this effect and improves the level of vitamin C and E as well as the activity of SOD, CAT, and glutathione peroxidase. This effect could be attributed to the protective effect of camel milk against ethanol-induced oxidative stress. Camel milk was reported to contain high concentrations of vitamins A, B2, C, and E [14]. Therefore, as the serum antioxidants are being consumed due to oxidation exerted by ethanol, they could be replenished and the levels were improved towards normal due to the camel milk administration. Certainly, as free radicals are generated in the ethanol-treated rats, these free radicals could be neutralized with the help of vitamin E and C present in camel milk resulting in the increased level of serum antioxidant vitamins and enzymes activity. It was earlier reported that an increase in SOD activity has been associated with ulcer healing [15], and a decrease in CAT activity is usually observed in gastric adenocarcinoma and *Helicobacter pylori*-infected stomach [16].

The results of this study also revealed that WBC, RBC, HBC, PCV, MCV, and PLT decreased significantly ($P < 0.05$) in ethanol-induced ulcer group compared to other groups. Treatment with camel milk and omeprazole counteract this effect and improve the level of hematological parameters toward normalcy. This may be attributed to the ability of camel milk to inhibit osmotic lysis of the red blood cells and also increase the antioxidant defense mechanism of the erythrocytes membrane [17]. Camel milk is rich in vitamin E [14], which is essential to rapid cell membrane repair, responsible for the prevention of cellular oxidation. Vitamin E helps in maintaining the flexibility of red blood cells which reduce the fragility and damage as a result of the oxidation of phospholipid membranes of the cells [18]. This vitamin also plays a vital role in red blood

cells formation in the bone marrow [19]. Vitamin E also raises the concentration of hemoglobin by increasing the activity of enzymes chain formation of hemoglobin, including the aminolevulinic acid (ALA) dehydratase as well as raise the readiness of some of the basic elements required in the production of hemoglobin; such as iron and copper [20]. These effects have a direct link to the increase in the PCV, RBC, and hemoglobin (HGB) observed in camel milk treated group. The increased in total WBC observed in the ulcer-induced untreated group may be connected to the role of WBC in body defense system against inflammatory processes occurring in the lining of the digestive tract. The lesion and inflammation of the digestive tract might have been relieved by the camel milk supplementation administration; this could be responsible for the decrease in WBC observed in camel milk and omeprazole treated groups. This finding is in concord with that of Al-Fartosi and Al-Adhadh [8].

The results of the histological examination revealed that the administration of ethanol results in the gastric lesion and moderately destroyed the architecture of stomach with a clear discontinuity in the mucosa (mild-moderate ulceration) in ethanol-induced untreated experimental rats (micrograph 1). Camel milk supplementation and omeprazole treatment resulted in healing of the stomach sections present as preserved transmural architecture with clearly defined mucosa, submucosa, and muscularis propria.

Conclusion

The change observed on antioxidant vitamins levels, antioxidant enzymes activity, and hematological indices, as well as stomach lesion in ethanol-induced rats, has been relieved due to camel milk administration. These lead to the conclusion that the milk may possess healing effects on an ethanol-induced model ulcer in rats. Therefore, camel milk supplementation could be employed as a means of managing ulcer patients.

References

[1] Klein LCJ, Gandolfi RB, Santin JR, Lemos M, Cechinel Filho V, de Andrade SF. Antiulcerogenic activity of extract, fractions, and some compounds obtained from *Polygala cyparissias* St. Hillaire & Moquin (Polygalaceae). Naunyn Schmiedebergs Arch Pharmacol 2010; 381(2):121–6.

[2] Konturek SJ, Konturek PC, Brzozowski T. Prostaglandins and ulcer healing. Review Article. J Physiol Pharmacol 2005; 56(5):5–31.

[3] Belaiche J. Observational survey of NSAID related upper gastro-intestinal adverse events in Belgium. Acta Gastroenterol Belgica 2002; 65(2):65–73.

[4] Satyanarayana MN. Capsaicin and gastric ulcers. Crit Rev Food Sci Nut 2006; 46(4):275–328.

[5] Knoess KH. Milk production of the dromedary. In: Proceeding of the IFS Symposium Camels, Sudan, pp 201–14, 1979.

[6] Mainasara AS, Isa SA, Dandare A, Ladan MJ, Saidu Y, Rabiu S. Blood pressure profile and insulin resistance in salt-induced hypertensive rats treated with Camel Milk. Mediterranean J Nutr Met 2016; 9:75–83.

[7] Dandare A, Zarewa AS, Abdullahi K, Rabiu S, Onwughara CA. Amilioretive effect of aqueous leaves extract of ziziphus mucronata in ethanol induced gastric ulcer model rats. J Appl Life Sci Int 2017; 15(2):1–8.

[8] Al-Fartosi KG, Al-Adhadh AJ. The protective role of camel's milk on some hematological parameters of male rats infected with gastric ulcer. World J Pharm Sci 2014; 2(11):1465–8.

[9] Mostofa R, Ahmed S, Begum M, Rahman S, Begum T, Uddin AS, et al. Evaluation of anti-inflammatory and gastric anti-ulcer activity of Phyllanthus niruri L. (Euphorbiaceae) leaves in experimental rats. BMC Complement Altern Med 2017; 17:267.

[10] Beers RF, Sizer IW. A spectrophotometric method for measuring the breakdown of hydrogen peroxide by catalase. J Bio Chem 1952; 195:133–40.

[11] Zhou YX, Zhang PY, Zhou RZ. The acute and subacute toxicity to ammonia on grass carp (Ctenopharyngodon idellus). Acta Hydrobiologica Sinica 1986; 10(1):32–8.

[12] Rutkowski M, Grzegorczyk K. Modifications of spectrophotometric methods for antioxidative vitamins determination convenient in analytic practice. Acta Sci Pol Technol Aliment 2007; 6(3):17–28.

[13] Tian Y, Jiang B, An L, Bao Y. Neuroprotective effect of catalpol against MPP+-induced oxidative stress in mesencephalic neurons. Eur J Pharmacol 2007; 568:142–8.

[14] Yousef MI. Aluminum-induced changes in hematobiochemical parameters, lipid peroxidation and enzyme activities of male rabbits: Protective role of ascorbic acid. Toxicology 2004; 199(1):47–57.

[15] Naito Y, Yoshikawa T, Ando T, Kishi A, Ueda S, Oyamada H, et al. Changes in superoxide dismutase activity in the gastric mucosa of peptic ulcer patients. J Clin Gastroenterol 1992; 14(1):131–4.

[16] Monari M, Foschi J, Calabrese C, Liguori G, Di FG, Rizzello F, et al. Implications of antioxidant enzymes in human gastric neoplasms. Int J Mol Med 2009; 24:693–700.

[17] Araki K, Rifkind J. The rate of osmotic hemolysis: a relation-ship with membrane bilayer fluidity. Biochim Biophys Acta 1981; 645(1):81–90.

[18] Kraus A1, Roth HP, Kirchgessner M. Supplementation with vitamin C, vitamin E or beta carotene influences osmotic fragility and oxidative damage of erythrocytes of zinc deficient rats. J Nutr 1997; 127(7):1290–6.

[19] McDowell L. Vitamins in animal and human nutrition. 2nd edition, Iowa State University Press, Iowa, IA, pp 155–225, 2000.

[20] Niki E, Noguchi N. Dynamics of antioxidant action of vitamin E. Acc Chem Res 2004; 37(1):45–51.