

Research Article

Evaluation of Gastroprotective Effect of Betalain-Rich Ethanol Extract from *Opuntia stricta* var. *Dillenii* Employing an In Vivo Rat Model

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The goal of this study is to investigate the antiulcer effects of betalain-rich extract (BRE). Gastric ulcer was induced by the administration of ethanol by gastric gavage route. This study showed that the supplementation of the BRE from pulp and peel at 800 mg/kg to rats with ethanol-induced gastric-ulcer significantly reduced the volume of gastric secretion (VGS) by 35% ($p = 0.001$) and 34% ($p = 0.0009$), the ulcer index (UI) by 41% ($p = 0.001$) and 68% ($p = 0.0008$), and the curative ratio (CR) by 41% and 68%, respectively, as compared to untreated ethanol-induced gastric ulcer. In addition, the administration of pulp and peel BRE to rats at dose 800 mg/kg significantly attenuates the variation in pH of gastric juice ($p = 0.008$ and $p = 0.001$) and its total acidity (TA) ($p = 0.001$ and $p = 0.001$). The antiulcer effect of BRE was confirmed by macroscopic and histological evaluation. Furthermore, pulp and peel BRE attenuated gastric ulcer-induced stress oxidants in rats' stomachs showed by a significant decrease in the lipid peroxidation rate ($p = 0.007$ and $p = 0.001$) and LDH activity ($p = 0.01$ and $p = 0.008$) and a potential increase in the superoxide dismutase (SOD) ($p = 0.01$ and $p = 0.008$), glutathione peroxidase (GPX) ($p = 0.006$ and $p = 0.001$), and catalase (CAT) ($p = 0.001$ and $p = 0.0009$). **Conclusion.** Therefore, this study shows for the first time that BRE, a natural colorant from *O. stricta*, is efficient in the amelioration of ulcer and stomach inflammation.

1. Introduction

Gastric ulcer disease is characterized by lesions and alterations in the gastric mucosa. Ulcers are characterized by anatomical destruction of the mucosa, which is immersed in gastric acid. This disease occurs as a result of many factors, such as a reduction in the mucus secretion associated with prolonged ingestion of anti-inflammatory drugs, infection with *Helicobacter pylori*, and others. Gastric ulcers are characterized by many symptoms such as gastric pain, vomiting, stress gastrointestinal symptoms, abdominal discomfort, and flatulence. In patients, with gastric ulcers, the treatment is performed using synthetic drugs such as proton pump inhibitors, antiacids, and H₂ receptor antagonists. Nevertheless, these synthetic drugs cause many toxic effects such as cardiac arrest and

liver-kidney toxicities [1–8]. Accordingly, recent research has focused on the search for effective plant-derived substances, which can be good sources for food additives as colorant, sweetener, preservative, and aroma [9–16]. Currently, there is a significant increase in the use of plant-derived colorants with a protective action against various perturbations and diseases such as diabetes, cancers, osteoporosis, and hyperlipidemia [17–32]. It is notable that the ingestion of *O. stricta* prickly pears also improves platelet function and hemostatic balance, thus contributing to prevent atherosclerotic risks [33, 34]. In this context, previous studies have reported that *Opuntia ficus-indica* consumption protects from gastric diseases and perturbation [8, 35–37]. Of special interest, betalains are plant water-soluble pigments that can be used as natural colorants in sweets, juices, ice creams, and beverages without toxic effects

[6, 7]. In fact, betalains have interesting technological properties such as solubility in water and food color intensity. Previous studies have reported that betalains prevent the oxidation of low-density lipoproteins (LDL) and therefore help reduce patients' cardiovascular disease risk. Moreover, betalains prevent inflammation in various organs such as the kidney, liver, and stomach [38, 39]. Previous studies have reported that betalains exhibit cancer proliferation in the human colon, ovaries, and cervical epithelium [39–44]. No study has, however, been provided on the antiulcer effect of betalain-rich extract. Accordingly, the present study evaluated the action of the administration of *O. stricta* betalain-rich extract from the pulp and peel to rats with ethanol-induced gastric ulcers by the gastric gavage route.

2. Materials and Methods

2.1. *O. stricta* Collection. Matured fleshy fruit *Opuntia stricta* was collected during the month of November 2021 without damage and were laved and skinned manually. The pulp or peel was blended and the seeds were removed and the juice was lyophilized.

2.2. Preparation of Betalain-Rich Extracts. The betalains from *O. stricta* (bar code ID: 000003165) pulp and peel were extracted according to the protocol of Sawicki et al. [45]. In brief, 20 g of pulp or peel from *O. stricta* and 10 mL of absolute methanol were mixed. The mixture was homogenized for 5 min. After, 1 g of sodium citrate, 0.01 g of disodium citrate, 1 g sodium chloride, and 4 g of magnesium sulfate were added. The mixture was homogenized for 5 min and then centrifuged for 15 min/3500 × *g*. Then, the supernatant was mixed with 0.15 g strong anion exchanger (SAX) sorbent and 1 g magnesium sulfate. The samples were homogenized for 3 min and centrifuged for 5 min/3500 × *g*. Finally, the supernatant was lyophilized, and two powders were obtained from the pulp and peel with betalain levels 94% and 96%, respectively.

2.3. Acute Oral Toxicity. To determine the toxic effect of BRE, various doses (0.2, 0.5, 0.8, and 1.6 g/kg, body weight) were administrated by the gastric gavage route [31, 46–48]. The general signs of toxicity hypoactivity, ventilation disorders, and mortality were verified daily over 3 days.

2.4. Antiulcer Activity Evaluation

2.4.1. Animal Model. 30 male *Wistar* rats weighing 165 ± 13 g and aged 7 weeks were housed under controlled environment and permitted water and food pellets ad libitum. The rats' experimentation conditions were approved by the Institutional Animal Ethics Committee (code: 86/609/EEC). Male *Wistar* rats were divided into four groups, six rats in each group. Control rats received distilled water at dose 0.5 ml/100g bw (C). Gastric ulcer rats received absolute ethanol at dose 0.5 mL/100 g · B.W by the gastric gavage route and were named U. U + 200, U + 400, and U + 800 BRE (pulp): gastric ulcer rats received 200 or 400 or 800 mg/kg bw from *O. stricta* pulp BRE. U + 200, U + 400, and U + 800 BRE

(peel): gastric ulcer rats received 200 or 400 or 800 mg/kg bw from *O. stricta* pulp peel BRE. U + 30 mg/kg OmepZ: ulcer rats received 30 mg/kg omeprazole as a synthetic proton pump inhibitor drug [49].

Absolute ethanol was administered by the gastric gavage route to rats, 1 hour after the BRE or omeprazole ingestion. 90 min later, the rats were sacrificed and the stomach was detached from rats and was opened along the greater curvature. The stomachs were gently rinsed with iced cold phosphate buffer for cleaning. The stomachs were strained and photos were taken to examine the gastric lesions. Parts of the stomach of the different groups of rats were crushed in phosphate buffer, after centrifugation at 5000 rpm, and the samples were stored at –80°C until used for the biochemical analysis. The area of gastric lesion was measured using ImageJ program.

2.4.2. Gastric Ulcerative Lesions and Ulceration Index Determinations. The total ulcer area for cleansed stomach was measured using an inverted microscope with a digital camera. The stomach ulcer area was calculated by the ImageJ software (version 1.51J8) with digital calculable distance (mm) by means of an e-ruler. The ulcer index (UI) was determined following the formula: ulcer index = ((ulcer area)/(total mucosa surface area)) × 100. The curative ratio was determined following the formula: curative ratio = (US control – US treated)/(US control) [50].

2.4.3. Analytical Methods. The gastric volume was calculated by the methods described by [51]. Gastric juice was collected by centrifugation of gastric mucus at 3500**g*/5 min/4°C to remove insoluble mucosal gastric. The gastric juice was calculated using graduate tubes. The stomach lipid peroxidation was estimated by the determination of the rate of thiobarbituric acid reactive substances (TBARS) by the technique described by Buege and Aust [52]. The superoxide dismutase activity was determined by the process of Marklund and Marklund [53]. The glutathione peroxidase (GPx) activity was determined according to the protocol of Pagila and Valentine [54]; and the catalase (CAT) activity was measured according to the protocol of Aebi [55]. The protein level was assayed by the determination of the level of proteins reacting with Cu²⁺ in the alkaline solution (Kits Biolabo, France, ref. 95010). The pH of the gastric juice was determined by a digital pH meter. The histological study was performed according to the process described in our previous studies [56].

2.5. Statistical Analysis. Tables and figures are presented as means ± standard deviation. The differences between all groups were evaluated using ANOVA and Fisher's test, and the significance variation was considered at *p* ≤ 0.02.

3. Results

3.1. Acute Toxicity Study. Rats supplemented with BRE at quantities 0.8 and 1.6 g/kg were reserved for 7 days. Rats supplemented with BRE at a dose that varies from 0.2 to 1.6 mg/kg were characterized by the absence of any signs of

toxicity and abnormalities such as behavioral changes, body weight changes, and biochemical analysis in blood changes.

3.2. Effects of BRE on Ethanol-Induced Macroscopic Lesions in Rats' Stomach. This study showed that BRE exerts a therapeutic effect against gastric ulcers induced by absolute ethanol. In fact, the administration of BRE from the pulp or peel at a dose of 800 mg/kg bw is protective against gastric macroscopic injury, which was shown by the presence of hemorrhagic ulceration and the decrease in UI by 41% and 68% ($p = 0.001$ and $p = 0.0008$) as compared to untreated ulcer rats. Furthermore, the animals treated with BRE at 800 mg/kg were protected from gastric mucosa injury and damage induced by ethanol, with a very similar aspect to that of the normal rats. In addition, the supplementation of BRE from the pulp induced a significant decrease in the ulcer score by 32%, 38%, and 41% at doses 200, 400, and 800 mg/kg, respectively ($p = 0.007$), as compared to untreated gastric ulcer rats. In gastric ulcer rats treated with BRE from peel, the reduction in ulcer score was 47%, 62%, and 68% ($p = 0.005$ and $p = 0.003$) after the administration of doses at 200, 400, and 800 mg/kg, respectively (Table 1; Figures 1 and 2).

3.3. Effects of BRE on VGS in Ethanol-Provoked Gastric Ulcer in Rats. Our study revealed that the gastric ulceration by ethanol induced an important rise in the VGS by 76% ($p = 0.005$) as compared to normal rats. The supplementation of BRE from pulp at doses 200, 400, and 800 mg/kg, however, decreased the gastric mucus secretion by 16%, 21%, and 24% ($p = 0.02$, $p = 0.002$, and $p = 0.001$) at doses 200, 400, and 800 mg/kg, respectively ($p = 0.02$), as compared to gastric ulcer untreated rats. In addition, the administration of BRE from peel augmented the VGS secretion by 21%, 27%, and 35% ($p = 0.01$, $p = 0.002$, and $p = 0.0009$) at doses 200, 400, and 800 mg/kg as compared to gastric ulcer in untreated ulcer rats (Table 1).

3.4. Effects of BRE on Gastric Juice pH and Total Acidity and in Ethanol-Induced Gastric Ulcer in Rats. The present study revealed that the administration of the BRE from the peel and pulp at different doses (200, 400, and 800 mg/kg) stopped the rise of the total acidity and increased the pH near normal rats. In addition, the highest doses of BRE from the pulp and peel have better antisecretory activity as evidenced by the augmentation of the pH by 33% and 44% ($p = 0.01$) and reduction in total acidity by 25% and 28% in pulp and peel BRE treatment as compared to the gastric ulcer in untreated rats (Table 1).

3.5. Stomach Antioxidant Enzyme Activities and Cell Index Toxicity in Ethanol-Provoked Gastric Ulcer. Table 2 indicates that ulcers caused a significant reduction in the SOD, CAT, and GPx activities by 35%, 78%, and 70% ($p = 0.009$ and $p = 0.005$), respectively, as compared to normal rats. In addition, the LDH activity and TBARS level in the stomach gastric ulcer in rats increased by 35% and 83% ($p = 0.01$ and

$p = 0.008$), respectively. The BRE pulp and peel administration at 800 mg/kg, however, increased the stomach SOD by 55% and 102% ($p = 0.009$ and $p = 0.007$), CAT by 75% and 100% ($p = 0.007$ and $p = 0.008$), and GPx activities by 58% and 104% ($p = 0.007$ and $p = 0.02$), respectively, as compared to untreated gastric ulcers in rats. Moreover, the stomach LDH activity and TBARS level were noted to decrease by 17% and 23% ($p = 0.01$ and $p = 0.02$) in gastric ulcers in rats treated with BRE from the pulp and by 22% and 41% ($p = 0.01$; $p = 0.007$) in gastric ulcers in rats treated with BRE from the peel, respectively.

3.6. Effects of BRE on Histological Evaluation of Gastric Damage. This study showed that supplementation of ethanol by the gastric gavage route caused various lesions in the stomach of rats such as severe disruption of the gastric mucosa, gastric mucosa flattening, and necrotic lesions penetrating deeply into the mucosal and submucosal layers as compared to the gastric mucosa of normal rats (Figure 3). In addition, we showed that the administration of BRE from the pulp and peel or omeprazole prevented the gastric mucosal ulcer, the flattening of gastric mucosa, and necrotic lesions in the stomachs of rats.

4. Discussion

Previous studies reported that *O. stricta* betalains characterized by the presence of four bioactive molecules, which are betanin, isobetanin and betanidin, and sorhamnetin-3-O-rutinoside in the pulp and peel [57]. It seems that treatment with betalains and betalain-rich diets is not only nontoxic but could also prove to be a promising alternative to supplement therapies in oxidative stress-, inflammation-, and dyslipidemia-related diseases such as stenosis of the arteries, atherosclerosis, hypertension, and cancer, among others [58]. The results of this study showed that feeding absolute ethanol to rats via gastric gavage caused acute stress in the stomach as evidenced by SOD, CAT, GPX, and LDH activities' depletion, as well as a significant increase in RBARS levels, resulting in gastric mucosal damage and alteration, hemorrhagic ulceration, and gastric mucosal flattening as gastric ulceration. In fact, this is a very common disease that causes mucosal damage and ulceration, as well as a decrease in mucus content, total acidity, progressive lesions, and damaged areas [17–25]. Furthermore, this study is the first to report that administration of natural food coloring such as betalain (200, 400, and 800 mg/kg) to ethanol-induced gastric ulcers in rats protected the gastric mucosa from injury, bleeding ulcers, gastric ulcers, and flattening of gastric mucosa. Furthermore, our study showed that 800 mg/kg BRE had the lowest UI compared to ulcerated rats treated with 200 and 400 BRE. Similarly, the results showed that gastric mucosal damage and histological changes were significantly reduced in rats treated with a BRE dose of 800 mg/kg bw compared to 200 and 400 mg/kg bw. This dose effect was also mentioned at UI, VGS, and TA levels. This study showed that taking 800 mg/kg body weight of BRE seemed to be better than lower doses. Onyeka et al.

TABLE 1: VGS and gastric juice pH and TA values and UI (%) and CR (%) in ethanol-induced gastric ulcer.

Groups	VGS (ml)	pH	TA	UI (%)	CR (%)
C	2.1 ± 0.4	5.7 ± 0.5	51 ± 5		
U	3.7 ± 0.2***	3.6 ± 0.3**	79 ± 4***	6.7 ± 0.8	
Pulp BRE					
BRE 200	3.1 ± 0.3***	4 ± 0.76*#	69 ± 6***	4.5 ± 0.7##@	32.8 ± 1.9
BRE 400	2.9 ± 0.5*#	4.2 ± 0.5*#	65 ± 5***	4.1 ± 0.7##@	38.8 ± 2.1
BRE 800	2.8 ± 0.6***	4.8 ± 0.6*##@	59 ± 6###@@	3.9 ± 0.5***	41.7 ± 2.3
Peel BRE					
BRE 200	2.91 ± 0.3*#	3.9 ± 0.6**	65.8 ± 3*#@	3.5 ± 0.3***	47.7 ± 3.1
BRE 400	2.76 ± 0.2*##	4.6 ± 0.7*##@	61 ± 8*#@	2.5 ± 0.3###@	62.6 ± 3.7
BRE 800	2.41 ± 0.8***	5.2 ± 0.3*##@@	57 ± 5###@@	2.03 ± 0.9###@@	68.9 ± 3.3
OmeprZ	2.85 ± 0.2*##	4 ± 0.27*#	72 ± 4.5***	3.67 ± 0.5***	45.2 ± 2.9

Each value represents the mean ± SEM for each group (n = 5). Notes. C: normal rats; U: gastric ulcerated rats; pulp BRE: gastric ulcerated rats treated with *O. stricta* pulp betalains at doses 200, 400, and 800 mg/kg bw; peel BRE: gastric ulcerated rats treated with *O. stricta* peel betalains at doses 200, 400, and 800 mg/kg bw; VGS: volume gastric secretion; T.A: total acidity; U.I: ulceration index; CR: curative ratio. The statistical analyses presented are as follows: * $p \leq 0.02$, ** $p \leq 0.01$, and *** $p \leq 0.001$ vs. normal rats; # $p \leq 0.02$, ## $p \leq 0.01$, and ### $p \leq 0.001$ vs. ethanol-induced gastric ulcer in rats. @ $p \leq 0.02$, @@ $p \leq 0.01$, and @@@ $p \leq 0.001$ vs. ethanol-induced gastric ulcer in rats treated with omeprazole.

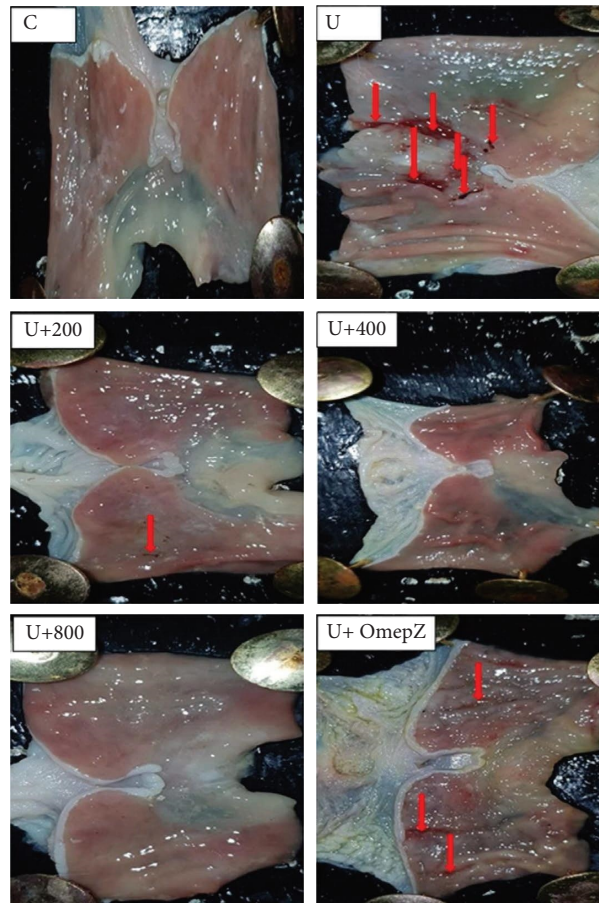


FIGURE 1: Photos of the pathological slides showing the effect of the administration of the pulp BRE on ethanol-induced gastric ulcer rats. C: normal appearance of the stomach without ulcer; U: gastric lesion and ulcerations with hemorrhagic regions. In ethanol-induced gastric ulcer rats treated with BRE from *O. stricta* pulp and peel at doses 200, 400, and 800 mg/kg bw (U + 200, U + 400, and U + 800), a protective effect from the gastric lesion was observed and only gastric mucosa are very milder compared to the injuries seen in the ulcer control rat. In omeprazole gastric ulcer rats (U + OmeprZ), a partial protective action from the gastric lesion and ulcerations with hemorrhagic regions was showed and this group presents few and middle hemorrhagic regions.

[36] showed that ingestion of the *Opuntia ficus-indica* extract in ethanol-treated rats protected against ethanol-induced gastric injury, ulceration, and histological

changes in the stomach, and the best protective effect was observed at a dose of 800 mg/kg body weight. This protective effect of BRE against ulcer and gastric damage is in

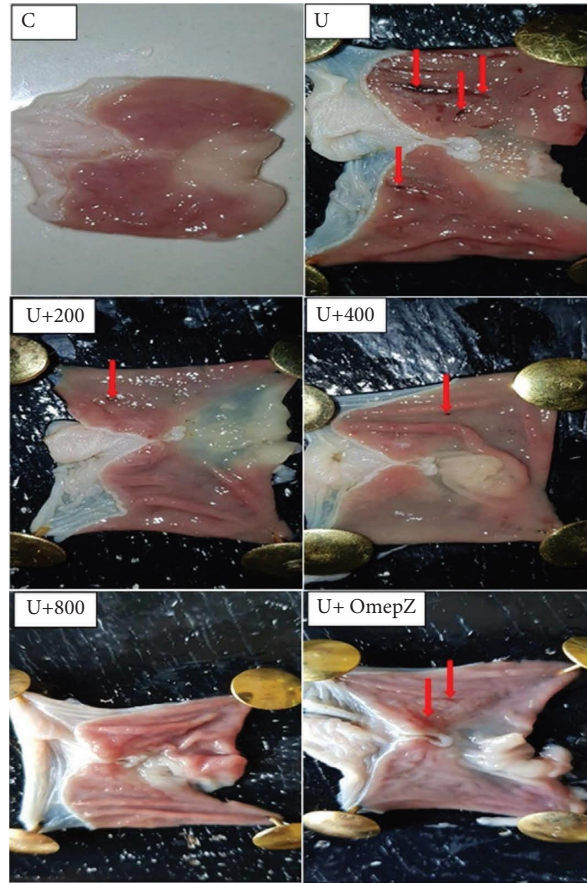


FIGURE 2: Photos of the pathological slides showing the effect of the administration of the peel BRE on ethanol-induced gastric ulcer rats. C: normal appearance of the stomach without ulcer; U: gastric lesion and ulcerations with hemorrhagic regions. In ethanol-induced gastric ulcer rats treated with BRE at doses 200, 400, and 800 mg/kg bw (U + 200, U + 400, and U + 800), an observable protective effect from ulceration of gastric mucosa and only a few and small lesions were showed as compared to the injuries seen in the ulcer control rat. In omeprazole gastric ulcer rats (U + OmeprZ), a partial protective action from the ulcerations with hemorrhagic of gastric mucosa was showed.

TABLE 2: Stomach SOD (U/g tissues), CAT ($\mu\text{moles H}_2\text{O}_2/(\text{min} \times \text{g tissues}/\text{min})$), GPx ($\mu\text{moles GSH}/(\text{min} \times \text{g tissues}/\text{min})$), and LDH (IU/mg protein) activities and TBARS levels in ethanol-induced gastric ulcer.

	C	U	U + 800 mg pulp BRE	U + 800 mg peel BRE	U + 30 mg/kg OmeprZ
SOD	0.79 ± 0.1	$0.34 \pm 0.02^{**}$	$0.53 \pm 0.14^{*#}$	$0.69 \pm 0.05^{##@}$	$0.51 \pm 0.2^{*#}$
CAT	3.6 ± 0.7	$1.6 \pm 0.6^{**}$	$2.8 \pm 0.11^{***##@}$	$3.2 \pm 0.4^{###@@}$	$1.8 \pm 0.3^{***#}$
GPx	22 ± 4	$12 \pm 2.3^{**}$	$19 \pm 1.2^{###}$	$24.5 \pm 4.9^{*###@}$	$16.1 \pm 2.5^{###}$
TBARS	0.61 ± 0.1	$1.12 \pm 0.3^{*#}$	$0.86 \pm 0.05^{*##}$	$0.65 \pm 0.07^{###@}$	$0.88 \pm 0.13^{##}$
LDH	21.5 ± 1.7	$29.1 \pm 2.1^{**}$	$24.1 \pm 2.3^{*#}$	$22.6 \pm 2.7^{##@}$	$26.8 \pm 3.2^{#}$

Notes. C: normal rats; U: gastric ulcerated rats; pulp BRE: gastric ulcerated rats treated with *O. stricta* pulp betalains at doses 200, 400, and 800 mg/kg bw; peel BRE: gastric ulcerated rats treated with *O. stricta* peel betalains at doses 200, 400, and 800 mg/kg bw; VGS: volume gastric secretion; T.A: total acidity; UI: ulceration index; CR: curative ratio. The statistical analysis is presented as follows: * $p \leq 0.02$, ** $p \leq 0.01$, and *** $p \leq 0.001$ vs. normal rats; # $p \leq 0.02$, ## $p \leq 0.01$, and ### $p \leq 0.001$ vs. ethanol-induced gastric ulcer in rats. @ $p \leq 0.02$, @@ $p \leq 0.01$, and @@@ $p \leq 0.001$ vs. ethanol-induced gastric ulcer in rats treated with omeprazole.

accordance with Babitha et al. [59], who showed that betalain protected against gastric mucosal lesions by inhibiting neutrophil infiltration and gastric ulceration. Kaur et al. [60] reported that betalains reduced neutrophil infiltration and protected the gastric mucosa from inflammation. Similarly, Hijazi et al. [61] reported that betalains increased NO levels and reduced proinflammatory

mediators, thereby protecting the gastric tissue from damage.

BRE protects the gastric mucosa from injury and bleeding ulcers leading to a reduction in the increase in total acidity, as indicated by an increase in the pH of gastric juice. Oxidative stress is thought to play a major role in gastric ulcers and lesions. In this study, in rats with ethanol-induced

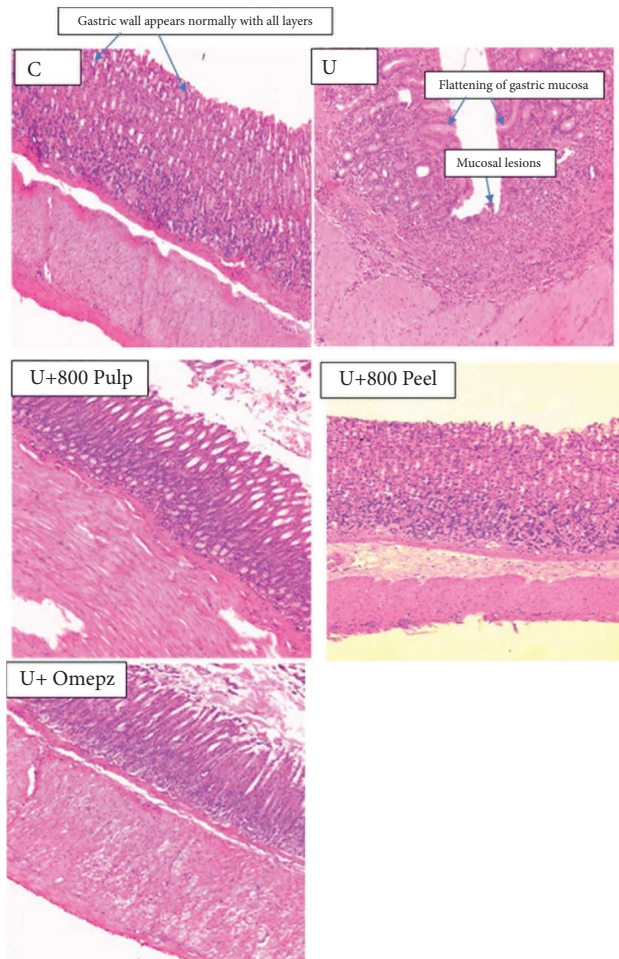


FIGURE 3: Photos of the pathological slides showing the effect of the administration of the pulp and peel BRE on ethanol-induced histological gastric mucosa ulcer in rat. There is severe disruption of the gastric mucosa, flattening of gastric mucosa, and necrotic lesions penetrating deeply into mucosal and submucosal layers in ulcer rats as compared to the gastric mucosa of normal rats (H&E stain, 10x). U + 800 mg BRE from pulp and U + 800 mg BRE from peel: gastric tissues are normal, mucus without ulceration, and only small and few lesions in gastric mucosa were observed. Stomach histological section of rats treated with OmeprZ at dose 30 mg/kg bw: mild disruption to the submucosal layer.

gastric ulcer, the activities of SOD, CAT, and GPx in the stomach of rats with gastric ulcer were significantly decreased, while the levels of TBARS and LDH were increased. However, administration of BRE to ethanol-induced gastric ulcer in rats stimulated the activities of gastric antioxidant enzymes such as SOD, CAT, and GPX. These enzymes are present in the human digestive tract and thus protect the gastric mucosa from damage and lesions, as showed by the increase in lipid peroxidation [36]. BRE stimulates the antioxidant system and protects the stomach from damage and lesions, and VGS was increased by 27% and 35% in gastric ulcers in rats treated with *O. stricta* pulp and peel natural pigments, respectively. The results of this study showed that gastric ulcers caused an increase in VGS compared with normal rats. In fact, one of the most important gastric ulcers

is the induction of gastric mucus secretion, an increase that may be a consequence of increased gastric acidity [62–64]. These results are in agreement with Hijazi et al. [61], who reported that beetroot betalains prevents mucosal damage by inhibiting neutrophil infiltration into the ulcerated gastric tissue. Similarly, Byeon et al. [65] studied the gastro-protective effect of beetroot pigment on various gastric ulcer models in rats. They showed that dose-dependent inhibition of UI and improvement of CR resulted in ethanol-induced gastric ulceration in rats after administration of BRE. Histological evaluation of the stomach revealed that gavage of absolute ethanol supplementation in rats resulted in severe gastric mucosal disruption, flattening of the gastric mucosa, and lesions deep into the mucosal and submucosal layers compared with normal rats. The anti-inflammatory activity of betalains has been reported to reduce inflammatory mediators and increase gastric anti-inflammatory cytokines [66]. Thus, betalains can be used in the prevention and treatment of gastric ulcers and injuries. In fact, natural pigments prevent tissue damage caused by ethanol and protect the mucosal layer from damage caused by ulcers [49, 67, 68]. Our results are in accordance with Ofusori et al. [50] who reported that *Celosia trigyna* pigments are protective against ethanol-induced gastric lesions and ulcers in mice. In addition, Martelli et al. [69] showed that natural food coloring as anthocyanins is a promising functional food for the treatment of gastric ulcers, gastrointestinal infection conditions, and upper gastrointestinal dyspepsia. Furthermore, Lakshmi [70] reported that feeding natural pigments derived from plants at a dose of 800 mg/kg bw reduced gastric ulcer surface area, as shown by the decrease of UI and the increase of CR; improved gastric ulcer-related biochemical indices such as VGS, TA, and pH; stimulated activity of gastric antioxidant enzymes such as SOD, CAT, and GPx; and reduced the lipid peroxidation. In another study, mice were supplemented with 100 mg/kg anthocyanin-rich food by the gastric gavage route to protect from mucosal gastric ulceration and damage, as compared to untreated rats [71]. In ulcerated rats, the administration of *Lycium barbarum* C-phycoerythrin significantly reduced the gastric cyclooxygenase-1, prostaglandin E2, and total nitrites and nitrate rates [68]. Galati et al. [72] evaluated the intestinal barrier protective activity of *O. stricta* juice against ulcers and gastric lesions. Previous studies reported that the administration of *Opuntia ficus-indica* juice to ethanol-induced gastric ulcer in rats increased gastric mucus production and protection of gastric mucosa against ulcers and lesions. Similarly, Kim et al. [73] showed that pigments from *Opuntia ficus-indica* at doses 800–1600 mg/kg significantly protect against gastric mucosal damage caused by stress.

5. Conclusion

The results of this study showed that *O. stricta* betalains from the pulp and peel protect against ethanol-induced macroscopic hemorrhagic damage in gastric mucosa, gastric mucosal decrease, and ulcer index UI. In addition, the supplementation prevented gastric mucosal damage as shown by an important increase in the gastric antioxidant

activity. In addition, this study suggested that the consumption of BRE at a dose of 800 mg/kg seemed to be better than the lower doses in order to prevent gastric mucosal damage, alteration, and inflammation caused by ethanol.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

Ethical Approval

All animals were used according to the guidelines of the Tunisian Society for the Care and Use of Laboratory Animals, and the study was approved by the University of Tunisia Ethical Committee (Code: 86/609/EEC). The Ethics and animal Experimentation Committee is made up of 30 members and chaired by Dr. Besma Bel Hadj Jrad (e-mail: bbhj2002@yahoo.fr).

Conflicts of Interest

The authors declare that there are no conflicts of interest.

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