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Full Length Research Paper

Gastroprotective effects of *Dicranopteris linearis* leaf extract against ethanol-induced gastric mucosal injury in rats

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Dicranopteris linearis **is a medicinal plant commonly used traditionally in the treatment of many ailments. This study was performed to evaluate the gastroprotective effect of ethanolic extracts of *D. linearis* leaf extract (DLELE) against ethanol-induced gastric mucosal injury in experimental rats. The rats were divided into four groups respectively pre-treated orally with carboxymethyl cellulose (CMC) solution (ulcer control groups), omeprazole 20 mg/kg (reference group), 250 and 500 mg/kg of DLELE (experimental groups) one hour before oral administration of absolute ethanol to generate gastric mucosal damage. After an additional hour, the rats were sacrificed and the ulcer areas of the gastric walls were determined. The ulcer control group exhibited severe mucosal injury, whereas groups pre-treated with DLELE exhibited significant protection of gastric mucosa. These findings were also confirmed by histology of gastric wall. Significant increases in gastric mucus production and decrease in acidity of gastric content were observed in treated groups with DLELE compare to ulcer control group. In conclusion, treatment with DLELE prior to absolute alcohol has significantly protect gastric mucosa as ascertained grossly by significant reduction of ulcer area, increases in gastric mucus production and decrease the acidity of gastric content and histology by comparatively decreases in gastric mucosal injury, reduction or absence of edema and leucocytes infiltration of submucosal layer compared to ulcer control group. DLELE was able to decrease the acidity and increase the mucosal defense in the gastric area, thereby justifying its use as an antiulcerogenic agent.**

Key words: *Dicranopteris linearis*, cytoprotection, gastric ulcer, mucus, histology.

INTRODUCTION

Gastric ulcer is an illness that affects a considerable number of people worldwide. The etiological factors of this disorder include stress, smoking, nutritional deficiencies, infections, frequent and indiscriminate use of nonsteroidal anti-inflammatory drugs (NSAIDs) (Khazaei and Salehi, 2006). The pathogenesis of gastroduodenal ulcers are influenced by various aggressive and defensive factors, such as mucus secretion, mucosal barrier, acid-pepsin secretion, blood flow, cellular regeneration and endogenous protective agents (prostaglandins and epidermal growth factor (Mizui et al., 1987). Although, the introduction of proton-pump inhibitors to the classic anti-ulcer therapy had revolutionized treatment of peptic ulcers and other gastrointestinal disorders, there is still no complete cure for this disease. It has been shown that long term use of these drugs leads to various adverse and side effects. Relapses of the malady, ineffectiveness of different drug

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regimens and even resistance to drugs are emerging (Al-Mofleh et al., 2007). Thus, there is an urgent requirement to identify more effective and safe anti-ulcer agents. A widespread search has been launched to identify new anti-ulcer therapies from natural sources. Herbs, medicinal plants, spices, vegetables and crude drug substances are considered to be a potential source to combat various diseases including gastric ulcer. In the scientific literature, a large number of medicinal plants with gastric anti-ulcer potential have been reported (Abdulla et al., 2010; Ketuly et al., 2011; Mahmood et al., 2010; Wasman et al., 2010).

Many plants are being used in the traditional medicine because they produce a diverse range of bioactive molecules, making them a rich source of different types of medicines (Tanaka et al., 2006). *Dicranopteris linearis* (Gleicheniaceae), known locally to the Malay's as Resam has been used in the Malay's traditional medicine as a cooling drink and also to reduce fever (Zakaria et al., 2006). In other part of the world, it is used to treat asthma and for women's sterility (Vasuda, 1999), and to get rid of intestinal worms infection (Chin, 1992). Scientifically, *D. linearis* extracts have been reported to possess anti-nociceptive, anti-inflammatory and antipyretic activities (Zakaria et al., 2008), antibacterial activity (Lai et al., 2009) and potential cytotoxic and antioxidant activity against various types of cancer (Zakaria et al., 2011). Phytochemical study has revealed the presence of various types of flavonoids, particuarly of flavonol 3-O-glycosides types, and triterpenes, saponins and high content of steroids in the leaves of *D. Linearis* (Raja et al., 1995; Zakaria, 2007). Thus far, there is no data available on gastroprotective activity of DLELE. The present study was undertaken to evaluate anti-ulcerogenic properties of DLELE in rats.

MATERIALS AND METHODS

In this study, omeprazole was used as the reference anti-ulcer drug, and was obtained from the University Malaya Medical Centre (UMMC) Pharmacy. The drug was dissolved in carboxymethyl cellulose (0.5% w/v) (CMC) and administered orally to the rats in concentrations of 20 mg/kg body weight (5 ml/kg) according to the recommendation of (Abdulla et al., 2010).

Plant specimen and extract preparation

D. linearis leaves were obtained from Ethno Resources Sdn Bhd, Selangor Malaysia, and identified by comparison with the Voucher specimen deposited at the Herbarium of Rimba Ilmu, Institute of Science Biology, University of Malaya, Kuala Lumpur. The dried leaves were powdered using electrical blender. Hundred grams of the fine powder were soaked in 500 ml of 95% ethanol in conical flask for 3 days. After 3 days the mixture was filtered using a fine muslin cloth followed by filter paper (Whatman No. 1) and distilled under reduced pressure in an Eyela rotary evaporator (Sigma-Aldrich, USA). The dry extract was then dissolved in CMC(0.5% w/v) and administered orally to rats in concentrations of 250 and 500 mg/kg body weight (5 ml/kg body weight) according to the recommendation of (Mahmood et al., 2010).

Experimental animals for gastric ulcer

Sprague Dawley healthy adult male rats were obtained from the Experimental Animal House, Faculty of Medicine, University of Malaya, and Ethic No. PM/27/07/2010/MAA (R). The rats were divided randomly into 4 groups of 6 rats each. Each rat that weighed between 200 - 225 g was placed individually in a separate cage (one rat per cage) with wide-mesh wire bottoms to prevent coprophagia during the experiment. The animals were maintained on standard pellet diet and tap water. The study was approved by the Ethics Committee for Animal Experimentation, Faculty of Medicine, University of Malaya, Malaysia. Throughout the experiments, all animals received human care according to the criteria outlined in the "Guide for the Care and Use of laboratory Animals" prepared by the National Academy of Sciences and published by the National Institute of Health.

Gastric ulcer-induction by absolute ethanol

The rats fasted for 48 h before the experiment (Abdulla et al., 2010), but were allowed free access to drinking water up till 2 h before the experiment. Gastric ulcer was induced by orogastric intubation of absolute ethanol (5 ml/kg) according to the method described by Mahmood et al. (2010). Ulcer control groups were orally administered vehicle (CMC, 0.5% w/v, 5 ml/kg). The reference group received oral doses of 20 mg/kg omeprazole in CMC (5 ml/kg) as positive control. Experimental groups were orally administered DLELE in CMC solution (5 ml/kg) at doses of 250 and 500 mg/kg . One hour after this pre-treatment all groups of rats were administered with absolute ethanol (5 ml/kg) in order to induce gastric ulcers (Abdulla et al., 2010). The rats were euthanized 60 min later (Ketuly et al., 2011) under an overdose of xylazin and ketamine anesthesia and their stomachs were immediately excised.

Measurement of mucus production

Gastric mucus production was measured in the rats that were subjected to absolute ethanol-induced gastric lesions. The gastric mucosa of each rat was obtained by gentle scraping the mucosa with a glass slide and the collected mucus were weighed by using a precision electronic balance (Ketuly et al., 2011; Wasman et al., 2010).

Measurement of acid content of gastric juice (pH)

Samples of gastric contents were analyzed for hydrogen ion concentration by pH metric titration with 0.1 N NaOH solutions using digital pH meter (Abdulla et al., 2010; Ketuly et al., 2011).

Gross gastric lesions evaluation

Ulcers of the gastric mucosa appear as elongated bands of hemorrhagic lesions parallel to the long axis of the stomach. Gastric mucosa of each rat was thus examined for damage. The length and width of the ulcer (mm) were measured by a planimeter (10 × 10 mm^2 = ulcer area) under dissecting microscope (×1.8). The ulcerated area was measured by counting the number of small squares, 2 mm × 2 mm, covering the length and width of each ulcer band. The sum of the areas of all lesions for each stomach was

Table 1. Effect of DLELE on ulcer area and inhibition percentage in rats.

Animal group	Pre-treatment (5 ml/kg dose)	Mucus production	pH of gastric content	Ulcer area $(mm)^2$ (Mean ± S.E.M)	Inhibition (%)
1	CMC (Ulcer control)	0.37 ± 0.01^a	3.88 ± 0.01^a	955.25 ± 2.82^a	-
2	Omeprazole (20 mg/kg)	0.57 ± 0.01^b	7.14 ± 0.33^b	148.3 ± 2.62^b	84.48
3	DLELE (250 mg/kg)	0.52 ± 0.01^c	5.6 ± 0.1^c	225.11 ± 3.65^c	76.43
4	DLELE (500 mg/kg)	0.77 ± 0.01^d	6.22 ± 0.01^c	38.08 ± 2.18^d	96.01

All values are expressed as mean ± standard error mean. Means with different superscripts are significantly different. The mean difference is significant at the p>0.05 level.

applied in the calculation of the ulcer area (UA) wherein the sum of small squares × 4 × 1.8 = UA (mm^2) according to the recommendation of Mahmood et al. (2010). The inhibition percentage (I.0%) was calculated using the following formula according to the recommendation of Wasman et al. (2010).

$(I\%) = [(UA_{control} - UA_{treated}) \div UA_{control}] \times 100\%$.

Histological evaluation of gastric lesions

Specimens of the gastric walls of each rat were fixed in 10% buffered formalin and processed in a paraffin tissue processing machine. Sections of the stomach were made at a thickness of 5 µm and stained with hematoxylin and eosin for histological evaluation (Abdulla et al., 2010; Ketuly et al., 2011).

Statistical analysis

All values were reported as mean ± S.E.M. Statistical significance of differences between groups was assessed using one-way ANOVA. A value of $p<0.05$ was considered significant.

RESULTS

pH of gastric content and mucus production

The acidity of gastric content in experimental animals pretreated with DLELE was decreased significantly compared to that of the ulcer control group ($p<0.05$). The mucus production of gastric mucosa also increases significantly ($p<0.05$) in animals pretreated with DLELE compared to the ulcer control group (Table 1).

Gross evaluation of gastric lesions

The anti-ulcer activity of DLELE in ethanol-induced gastric lesion model is shown in Table 1. Results showed that rats pre-treated with DLELE extracts before being given absolute alcohol had significantly reduced areas of gastric ulcer formation compared to rats pre-treated with CMC (ulcer control group) (Figure 1) ($p<0.05$). Moreover, the DLELE significantly suppressed the formation of the ulcers and it was interesting to note the flattening of gastric mucosal folds in rats pretreated with 500 mg/kg DLELE (Figure 1). Furthermore, ethanol-induced mucosal damage was significantly and dose dependently reduced in the size and severity by pretreatment of the animals with DLELE. The significant inhibition of gastric ulcer in pretreatment with DLELE was comparable with omeprazole which is a standard drug used for curing gastric ulcer.

Histological evaluation of gastric lesions

Histological observation of ethanol induced gastric lesions in ulcer control group pre-treated with CMC only, showed comparatively extensive damage to the gastric mucosa, and edema and leucocytes infiltration of the submucosal layer (Figure 2). Rats that received pre-treatment with DLELE had comparatively better protection of the gastric mucosa as seen by reduction in ulcer area, reduced or absent submucosal edema and leucocytes infiltration (Figure 2). The DLELE has been shown to exert the cytoprotective effects in a dose-dependent manner.

DISCUSSION

Results obtained in current study suggest that DLELE administered at the low and higher dose showed a protective action against ethanol-induced gastric mucosa damage as demonstrated by the reduction of the gastric ulcer area, increased gastric mucous production and decreased the acidity of gastric content. DLELE prevented ethanol induced-gastric damage with mucous production increase. This may be explained with a correlation to a strengthening of the defence factors of gastric mucosa. It is evident that increased mucus production must have largely contributed to preventive effect of the DLELE. Similar findings exist in the literatures, where plant extracts have been shown to prevent gastric mucosal ulceration in rats (Ketuly et al., 2011; Wasman et al., 2010). The mucus of the gastric wall is thought to play an important role as a defensive factor against gastrointestinal damage (Wasman et al., 2010). Pretreatment with DLELE significantly decreases the acidity of the gastric content and increases the gastric

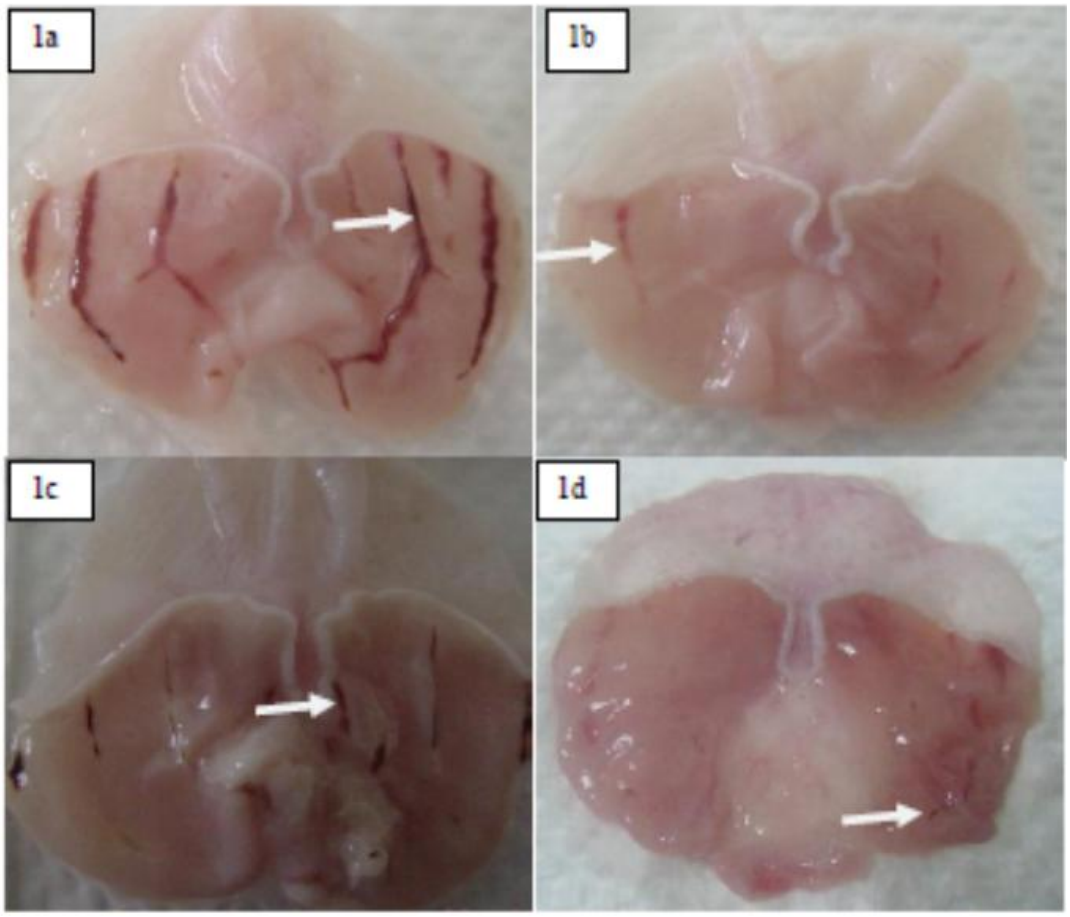

Figure 1. Gross appearance of the gastric mucosa in rats. (a) pre-treated with 5 ml/kg CMC (ulcer control). Severe injuries are seen in the gastric mucosa (arrow). Absolute ethanol produced extensive visible hemorrhagic necrosis of gastric mucosa. (b) pre-treated with of omeprazole (20 mg/kg). Injuries to the gastric mucosa are very milder compared to the injuries seen in the ulcer control rats (arrow). (c) pre-treated with DLELE (250 mg/kg). Mild injuries are seen in the gastric mucosa. The extract reduces the formation of gastric lesions induced by absolute ethanol (arrow). (d) pre-treated with DLELE 500 mg/kg. Mild injuries to the gastric mucosa are seen, and flattening of the gastric mucosa is seen (arrow).

mucus production. This suggests that gastro-protective effect of DLELE is mediated partly by preservation of gastric mucus production.

Oxidative stress plays an important role in the pathogenesis of various diseases including gastric ulcer, with antioxidants being reported to play a significant role in the protection of gastric mucosa against various necrotic agents (Trivedi and Rawal, 2001). Administration of antioxidants inhibits ethanol-induced gastric injury in rat (Ligumsky et al., 1995). DLELE possesses a broad spectrum of biological activities, and the plant extract has been shown to contain pharmaceutically active chemical constituents, such as flavonoids, saponins and terpenoids (Zakaria, 2007) and it is speculated that the gastroprotective effect exerted by DLELE could be attributed to its antioxidant property. Antioxidant property of the DLELE may possibly counteract oxidative damage caused by absolute ethanol toxicity. The observed anti-ulcerogenic activity may be due to its antioxidant effects and appears to strengthen the mucosal barrier, which is the first line of defense against endogenous and exogenous ulcerogenic agents. Previous studies have shown that flavonoids may be related to the antiulcer activity (Hiruma-Lima et al., 2006), and play a major role in the mechanism of gastroprotection (La Casa et al., 2000). It could be conceivable that the anti-ulcer activity of this plant could be linked to the flavonoids since flavonoids are reported to protect the mucosa by preventing the formation of lesions by various necrotic agents (Saurez et al., 1996). It is well known that many flavonoids display anti-secretory and cytoprotective properties in different experimental models of gastric ulcer (Zayachkivska et al., 2005). Flavonoids possess anti-oxidant properties in addition to strengthening the mucosal defense system through stimulation of gastric mucus secretion (Martin et al., 1994) and flavonoids can

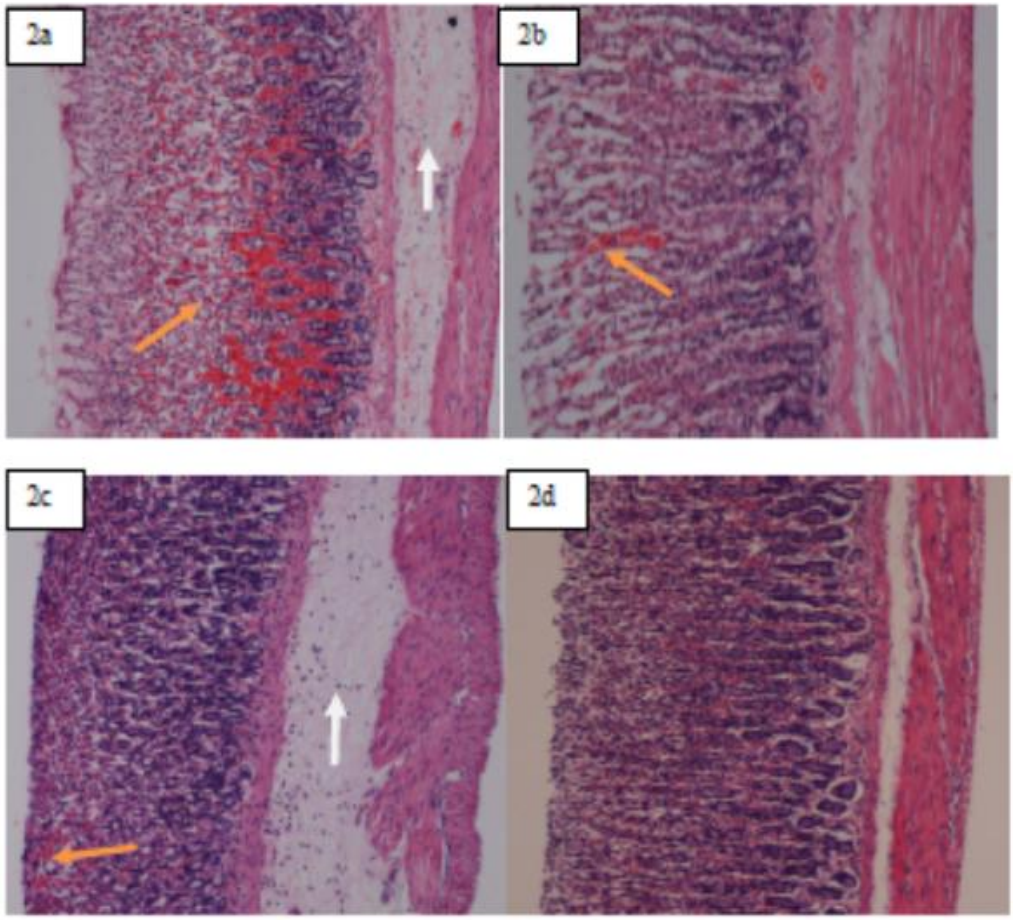

Figure 2. Histological study of the absolute ethanol-induced gastric mucosal damage in rats (a) pre-treated with 5 ml/kg of CMC (ulcer control). There is severe disruption to the surface epithelium, necrotic lesions penetrate deeply into mucosa (orang arrow) and extensive edema of submucosa layer and leucocyte infiltration are present (White arrow). (b) pre-treated with omeprazole (20 mg/kg). Mild disruption of the surface epithelium mucosa are present but deep mucosal damage is absent. (c) pre-treated with DLELE (250 mg/kg). Moderate disruption of surface epithelium are present but deep mucosal damage is absent. There is edema and leucocytes infiltration of the submucosal layer. (d) pre-treated with DLELE (500 mg/kg). There is no disruption to the surface epithelium with no edema and no leuco.

scavenge for the reactive oxygen species (super-oxide anions) and free radicals produced by ethanol. These reactive intermediates are potentially implicated in ulcerogenicity (Lewis and Hanson, 1991).

It is generally known that the antioxidant activities of putative antioxidants involves various mechanisms, such as radical scavenging, decomposition of peroxides, binding of transition metal ion catalysts, prevention of chain initiation and of continued hydrogen abstraction (Diplock et al., 1998). Hence, the free radical scavenging capacity of an extract may serve as a significant indicator of its potential antioxidant activity. Increasing evidences have suggested that many age-related human diseases are the result of cellular damage caused by free radicals (Carr and Frei, 2000), Antioxidants have been shown to play an important role in preventing such diseases. For example, several cancer chemopreventive agents exhibit antioxidant activity through their ability to scavenge oxygen radicals (Ito et al., 1999). DLELE demonstrated to contain flavonoids, saponins, triterpenes, tannins and steroids (Zakaria, 2007). The interests in phenolic compounds, particularly flavonoids and tannins, have considerably increased in recent years because of their broad spectrum of chemical and diverse biological properties, which include the antioxidant effects (Larson, 1988) and radical scavenging properties (Agrawal, 1989). Flavonoids have been associated with possible role in the prevention of several chronic diseases involving oxidative stress (Lee et al., 2003), as well as their protective effect against low-density lipoprotein (LDL) oxidation (Silva et al., 2000). Flavonoids have been reported to inhibit cytokine (inflammatory stimuli) release from RAW264.7

cells (Xagorari et al., 2002) and may modulate the increasing number of cellular processes involving redox reaction, including the regulation of tyrosine phosphatase activity (Gamet-Payrastre et al., 1999).

The result of the present study also revealed protection of gastric mucosa and inhibition of leucocytes infiltration of gastric wall in rats pretreated with DLELE. DLELE have been shown to contain anti-inflammatory activity (Zakaria et al., 2006) and it is speculated that the gastroprotective effect exerted by this plant extract could be attributed to its anti-inflammatory activity. This anti-inflammatory activity could also be a key factor in the prevention of gastric ulcer as reported by Swarnakar et al. (2005). Similarly, Abdulla et al. (2010) and Wasman et al. (2010) demonstrated that the reduction of neutrophil infiltration into ulcerated gastric tissue promotes the healing of gastric ulcers in rats. Mahmood et al. (2010) and Wasman et al. (2010) showed that oral administration of plant extract before ethanol administration significantly decreased neutrophil infiltration of gastric mucosa. Absolute alcohol would extensively damage the gastric mucosa leading to increased neutrophil infiltration into the gastric mucosa. Oxygenfree radicals derived from infiltrated neutrophils in ulcerated gastric tissues have inhibitory effect on gastric ulcers healing in rats (Suzuki et al., 1998). Neutrophils mediate lipid peroxidation through the production of superoxide anions (Zimmerman et al., 1997). Neutrophils are a major source of inflammatory mediators and can release potent reactive oxygen species such as superoxide, hydrogen peroxide and myeloperoxidase derived oxidants. These reactive oxygen species are highly cytotoxic and can induce tissue damage (Cheng and Koo, 2000). Furthermore, neutrophil accumulation in gastric mucosa has been shown to induce gastric ulceration (Abdulla et al., 2010; Wasman et al., 2010; Ketuly et al., 2011). Suppression of neutrophil infiltration during inflammation was found to enhance gastric ulcer healing (Mahmood et al., 2010). Studies have demonstrated the link between the anti-inflammatory and antioxidant activities of the plants. For example, nitric oxide (NO) is produced/ released under the action of inflammatory stimuli (that is, ROS) (Olszanecki et al., 2002). Inhibition of ROS leads to the reduction of NO production, which has been demonstrated to cause anti-inflammatory and antioxidant activities (Middleton et al., 2000). The free radical scavenging property may be one of the mechanisms by which these plants' are effective in their ethnopharmacological uses against different ailments.

In the present study, we observed flattening of the mucosal folds which suggests that gastroprotective effect of DLELE might be due to a decrease in gastric motility. It is reported that the changes in the gastric motility may play a role in the development and prevention of experimental gastric lesions (Abdulla et al., 2010; Ketuly et al., 2011). Relaxation of circular muscles may protect the gastric mucosa through flattening of the folds. This will increase the mucosal area exposed to necrotizing agents and reduce the volume of the gastric irritants on rugal crest (Mahmood et al., 2010; Wasman et al., 2010). Ethanol produces a marked contraction of the circular muscles of rat fundic strip. Such a contraction can lead to mucosal compression at the site of the greatest mechanical stress, at the crests of mucosal folds leading to necrosis and ulceration (Abdulla et al., 2010).

Conclusion

The study reveals DLELE could significantly protect the gastric mucosa against ethanol-induced injury. Such protection was ascertained grossly by increased gastric mucus production, decrease in the acidity of gastric content were significantly higher in treated groups compare to ulcer control group and also the reduction of ulcer areas in the gastric wall as well as histology by the reduction or inhibition of edema and leucocytes infiltration of submucosal layers. The data obtained confirm the traditional indications for this herb and present a new therapeutic option for the treatment of gastric ailments. The exact mechanism (s) underlying this anti-ulcerogenic effect remain unknown, but it seems that this extract contains pharmacologically active substances with potent antioxidant and anti-inflammatory activity which increase the mucus production and decrease the acidity of gastric content.

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REFERENCES

Abdulla MA, Ahmed KAA, AL-Bayaty, FH, Masood, Y (2010). Gastroprotective effect of *Phyllanthus niruri* leaf extract against ethanol-induced gastric mucosal injury in rats. Afr. J. Pharm. Pharmacol., 4(5): 226-230.

Agrawal PK (1989). Carbon-13 NMR of Flavonoids. New York: Elsevier Press, pp. 5-31.

Al-Mofleh IA, Alhaider AA, Mossa JS, Al-Soohaibani MO, Rafatullah S (2007). Aqueous suspension of anise *Pimpinella anisum* protects rats against chemically induced gastric ulcers. World J. Gastroenterol., 13: 1112-1118.

Carr A, Frei B (2000). The role of natural antioxidants in preserving the biological activity of endothelium-derived nitric oxide. Free Rad. Biol. Med., 28: 1806-1814.

Cheng CL, Koo MWL (2000). Effect of *Centella asiatica* on ethanol induced gastric mucosal lesions in rats. Life Sci., 67: 2647-2653.

Chin WY (1992). A Guide to Medicinal Plants. Singapore Sci. Centre, p. 24.

Diplock AT, Charleux JL, Crozier-Willi G, Kok FJ, Rice-Evans C, Roberfroid M, Stahl W, Vina-Ribes J (1998). Functional food science and defence against reactive oxygen species. Br. J. Nutr., 80: S77-S112.

Gamet-Payrastre L, Manenti S, Gratacap MP, Tulliez J, Chap H, Payrastre B (1999). Flavonoids and the inhibition of PKC and PI 3-kinase. Gen. Pharmacol., 32: 279-286.

Hiruma-Lima CA, Calvo TR, Rodrigues CM, Andrade FD, Vilegas W, Brito AR (2006). Antiulcerogenic activity of *Alchornea castaneafolia*: effects on somatostatin, gastrin and prostaglandin. J. Ethnopharmacol., 104: 215-224.

Ito H, Miyake M, Nishitani E, Mori K, Hatano T, Okuda T, Konoshima T, Takasaki M, Kozuka M, Mukainaka M, Tokudo H, Nishino H, Yoshida T (1999). Antitumor promoting activity of polyphenols from Cowania mexicana and Coleogyne ramosissima. Cancer Lett., 143: 5-13.

Khazaei M, Salehi H (2006). Protective effect of falcaria vulgaris extract on ethanol induced gastric ulcer in rat. Iran J. Pharmacol. Ther., 5: 43-46.

Ketuly KA, Abdulla MA, Hadi HA, Mariod AA, Abdel-Wahab SI (2011). Anti-ulcer activity of the 9alpha-bromo analogue of *Beclomethasone dipropionate* against ethanol-induced gastric mucosal injury in rats. J. Med. Plants Res., 5(4): 514-520.

La Casa C, Villegas I, Alarcon de la Lastra C, Motilva V, Martin Calero MJ (2000). Evidence for protective and antioxidant properties in rutin, a natural flavones, against ethanol induced gastric lesions. J. Ethnopharmacol., 71: 45-53.

Lai HY, Lim YY, Tan SP (2009). Antioxidative, tyrosinase inhibiting and antibacterial activities of leaf extract from medicinal ferns. Biosci. Biotechnol. Biochem., 73(6): 1362-1366.

Larson RA (1988). The antioxidants of higher plants. Phytochem., 27: 969-978.

Lee KW, Kim YJ, Lee HJ, Lee CY (2003). Cocoa has more phenolic phytochemicals and a higher antioxidant capacity than teas and red wine. J. Agric. Food Chem., 51: 7292-7295.

Lewis DA, Hanson PJ (1991). Antiulcer drugs of plant origin. Prog. Med. Chem., 28: 210-229.

Ligumsky M, Sestieri M, Okon F, Ginsburg I (1995). Antioxidants inhibit ethanol-induced gastric injury in the rat. Role of manganese, glycin and carotene. Scand. J. Gastroenterol., 30: 854-860.

Olszanecki R, Gêbska A, Kozlovski VI, Grylewski RJ (2002). Flavonoids and nitric oxide synthase. J. Physiol. Pharmacol., 53: 571-584.

Mahmood AA, Mariod AA, Al-Bayaty F, Abdel-Wahab SI (2010). Anti-ulcerogenic activity of *Gynura procumbens* leaf extract against experimentally-induced gastric lesions in rats. J. Med. Plants Res., 4(8): 685-691.

Martin MJ, Marhuenda E, Perez-Guerrero C, Franco JM (1994). Antiulcer effect of naringin on gastric lesion induced by ethanol in rats. Pharmacology, 49: 144-150.

Middleton Jr. E, Kandaswami C, Theoharides TC (2000). The effects of plant flavonoids on mammalian cells: implications for inflammation, heart disease, and cancer. Pharmacol. Rev., 52: 673-751.

Mizui T, Sato H, Hirose F, Doteuchi M (1987). Effect of antiperoxidative drugs on gastric damage induced by ethanol in rats. Life Sci., 41: 755-763.

Raja DP, Manickam VS, De Britto AJ, Gopalakrishnan S, Ushioda T, Satoh M, Tanimura A, Fuchino H, Tanaka N (1995). Chemical and chemotaxonomical studies on Dicranopteris species. Chem. Pharm. Bull., 43: 1800-1803.

Saurez J, Herreta MD, Marhuenda E (1996). Hesperidine and neohesperidine dihydrochalcone on different experimental models of induced gastric ulcer. Phytother. Res., 10: 616-618.

Silva FA, Borges F, Guimaraes C, Lima JL, Matos C, Reis S (2000). Phenolic acids and derivatives: Studies on the relationship among structure, radical activity, and physicochemical parameters. J. Agric. Food Chem., 48: 2122-2126.

Suzuki Y, Ishihara M, Ito M (1998). Anti-ulcer effects of antioxidants, quercetin, α-tocopherol, nifedipine and tetracycline in rats. Jpn. J. Pharmacol., 78: 435-441.

Swarnakar S, Ganguly K, Kundu P, Banerjee A, Maity P, Sharma AV (2005). Curcumin regulates expression and activity of matrix metalloprotinases 9 and 2 during prevention and healing of indomethacin-induced gastric ulcer. J. Biol. Chem., 280: 9409-9415.

Tanaka JCA, Da Silva CC, De Oliveira AJB, Nakamura CV, Dias Filho BP (2006) Antibacterial activity of indole alkaloids from *Aspidosperma ramiflorum*. Braz. J. Med. Biol. Res., 39: 387-391

Trivedi NP, Rawal UM (2001). Hepatoprotective and antioxidant property of *A. paniculata* Nees in BHC induced liver damage in mice. Indian J. Exp. Biol., 39: 41-46.

Vasuda SM (1999). Economic importance of pteridophytes. Indian Fern. J. 16: 130-152.

Wasman SQ, Mahmood AA, Salehhuddin H, Zahra AA, Salmah I (2010). Cytoprotective activities of *Polygonum minus* aqueous leaf extract on ethanol-induced gastric ulcer in rats. J. Med. Plants Res., 4(24): 2658-2665.

Xagorari A, Roussos C, Papapetrpoulos A (2002). Inhibition of LPSstimulated pathways in macrophages by the flavonoid luteolin. Br. J. Pharmacol., 136: 1058-1064.

Zakaria ZA (2007). Free radical scavenging activity of some plants available in Malaysia. Iran J. Pharmacol. Ther., 6: 87-91.

Zakaria ZA, Abdul Ghani ZDF, Raden Mohd Nor RNS, Hanan Kumar G, Sulaiman MR, Fatimah CA (2006). Antinociceptive and antiinflammatory activities of *Dicranopteris linearis* leaves chloroform extract in experimental animals. Yakugaku Zasshi, 126: 1197-1203.

Zakaria ZA, Ghani ZD, Nor RN, Gopalan HK, Sulaiman MR, Jais AM, Somchit MN, Kader AA, Ripin J (2008). Antinociceptive, anti-inflammatory, and antipyretic properties of an aqueous extract of *Dicranopteris linearis* leaves in experimental animal models. J. Nat. Med., 62(2):179-87.

Zakaria ZA, Mohamed AM, Mohd. Jamil NS, Rofiee MS, Somchit MN, Zuraini A, Arifah AK, Sulaiman MR (2011). *In vitro* cytotoxic and antioxidant properties of the aqueous, chloroform and methanol extracts of *Dicranopteris linearis* leaves. Afr. J. Biotechnol., 10(2): 273-282.

Zayachkivska OS, Konturek SJ, Drozdowicz D, Konturek PC, Brzozowski T, Ghegotsky MR (2005). Gastroprotective effects of flavonoids in plant extracts. J. Physiol. Pharmacol., 56: 219-231.

Zimmerman JJ, Ciesielski W, Lewandoski J (1997). Neutrophil-mediated phospholipids peroxidation assessed by gas chromatography-mass spectroscopy. Am. J. Physiol., 273: 653-661.