

Gastro-protective Effects of Honey in Male Wistar Rats

Owoyele B.V.,¹ *Ayinla, M.T.,¹ Esan, A.A.¹ and Bayo-Olugbami A²

¹Department of Physiology, Faculty of Basic Medical Sciences, University of Ilorin, Ilorin, PMB 1515, Nigeria
Page | 4114 ²Neuroscience Unit, Department of Physiology, Faculty of Basic Medical Sciences, Adeleke University, Ede, Osun
State, PMB 250, Nigeria

Date Received: 01-12-2020

Date Accepted: 28-05-2021

DOI: <https://doi.org/10.48198/NJPAS/20.B22>

ABSTRACT

Honey is consumed as food and also used in the treatments of ailment. However, honey of various types exhibit varying properties. Their therapeutic effects are determined by whether the honey is multifloral or monofloral, and also on the variety of nectars the honey is derived from. Manuka and Tualang types of honey were observed to possess anti-ulcer effects. However, no such report is available for University of Ilorin honey. This study therefore aimed to determine the gastroprotective effects of University of Ilorin honey in Wistar rats using two laboratory models for inducing ulcer (HCl/Ethanol and Indomethacin). Twenty rats each were assigned to four groups for each model of ulcer. The rats were treated for 12 days with the administration of distilled water (control), cimetidine (100 mg/kg b.w.) and University of Ilorin honey (250 mg/kg b.w. in one group and 1000 mg/kg b.w. in another group). The rats were fasted for 24 hours after the last treatment. Then, the ulcerogens were given orally. After four hours, the rats were sacrificed, gastric juice was collected and the stomachs were examined for ulceration. The results revealed that pre-treatment with University of Ilorin honey reduced gastric index from 6.80 ± 0.20 (1000 mg/kg b.w. to 2.40 ± 0.24 (control) $P < 0.05$ and 7.20 ± 0.37 (1000 mg/kg b.w.) to 3.20 ± 0.37 (control) $P < 0.05$ in HCl/Ethanol and indomethacin induced ulcer models respectively. There was no significant difference in ulcer index between the cimetidine administered rats and the honey administered group (1000 mg/kg bw.), but the honey group (250 mg/kg bw.) was less effective than the cimetidine group in the models used. Also, there were no changes in gastric acidity in all the groups compared with the control group. In conclusion, the results showed that University of Ilorin honey has protective effect on the gastrointestinal tract which is not due to alteration of gastric acidity.

Keywords: Honey, Gastric acidity, Indomethacin, Cimetidine, Ulcer index

Introduction

An ulcer is a lesion/erosion on the epithelial mucosa that extends through the muscularis mucosa into the submucosa or deeper (Khazael and Salehi, 2006). It occurs when aggressive and

mucosal irritating factors like lipid peroxidation, pepsin and acid secretion, overwhelm defensive factors such as mucous secretion, prostaglandins, glycoproteins and anti-oxidant enzymes (catalases,

Corresponding Author: Ayinla M.T.

Department of Physiology, University of Ilorin, Ilorin, Nigeria.

Phone: +2348076029175; Email: gazmark@unilorin.edu.ng



superoxide dismutase, glutathione) (Prabha *et al.*, 2009; Sung *et al.*, 2009).

The major causative agent of ulcer is a bacteria called *Helicobacter pylori* that colonize the antral mucosa. The bacteria cause a chronic active gastritis which result in defective regulation of gastrin production by the G cells in the stomach. Non-steroidal anti-inflammatory drugs (NSAIDs) is another major cause of peptic ulcer. It blocks the function of cyclooxygenase I, an enzyme essential in the production of prostaglandins. (Amandeep *et al.* 2012; Sharifi-Rad *et al.*, 2018). In the developed countries, peptic ulcer has a prevalence of 40% while a prevalence of 80% is found in developing countries (Goyal, 2008; Malfertheiner, 2009).

Ulcer is associated with increased morbidity and mortality for decades, not too long ago, epidemiological trends reveal an impressive fall in its incidence (Losano, 2012). Among the vital developments which led to reduced incidence of ulcer disease is the discovery of effective and potent acid suppressant. Despite this significant progress, peptic ulcer disease still constitutes a major morbidity as a result of extensive use of NSAID (Malfertheiner, 2009).

Although many medications are currently employed to manage ulcer including antacids, histamine H₂ receptor antagonist and proton pump inhibitors. However, they often cause many side effects. Hence the need to discover newer drugs that are safe, effective, and affordable and with minimal side effects. Hence, this study investigated an alternative source for the prevention of ulcer with the use of common dietary sources as a gastro-protective agent.

Honey is a sticky golden brown fluid that has a sweet taste and is produced by bees from the nectar of flower. Components of honey are carbohydrates, free amino acids, vitamins, trace elements and phenolic compounds e.g. flavonoids (Kassim *et al.*, 2010). Flavonoids are the most abundant compound found in honey and are closely involved in its biological functions (Alvarez-suarez *et al.*, 2013). Honey is added as sweetener for food,

medically it is also used for wound dressing and associated inflammatory conditions (Molan, 2001; Biglari *et al.*, 2013).

Other therapeutic benefits of honey include antimicrobial, antioxidants and analgesics (Azim *et al.*, 2007; Alzubier and Okechukwu, 2011; Biglari *et al.*, 2013; Gunduz *et al.*, 2013). Although honey production mechanism by bees is similar all over the World, the chemical and physical characteristics of honey varies as a result of different geographical and botanical origins (Akharaiyi and Lawal, 2016).

These differences in chemical composition, nutritional values and flavour of honey indicate that it is produced from different floral sources, mainly from plants (Omoya and Akharaiyi, 2010). Similarly, studies have shown that the quality of honey is determined by its physicochemical or biological properties specified in Codex Alimentarius standards (Bogdanov *et al.*, 2004), such qualities include moisture, ash, pH, reducing and non-reducing sugar. Hence, there is Malaysian (Gelam) honey (Kassim *et al.*, 2012), Acacia Honey (Azim *et al.*, 2007), University of Ilorin Honey/Unilorin honey etc. This research focuses on Unilorin honey which is a multifloral honey derived from nectars of local plants (Abdulrahman *et al.*, 2013). Unilorin honey was reported to have a moisture content of 18.20%, ash (0.2 %) (ash indicate mineral composition of honey), pH of 4.2 and total sugar content of 76.10% the latter has 74.00% as reducing sugar and 2.10% as non-reducing sugar (Okeola *et al.*, 2015). In addition, it contains potassium (29.25 mg/L), calcium (4.13 mg/L), iron (0.91 mg/L), copper (0.64 mg/L) and Zinc (2.94 mg/L). All these values fall within the Codex Alimentarius standards (Okeola *et al.*, 2015). Its different constituents provide both nutritional and health benefits (Okeola *et al.*, 2015; Ekanem *et al.*, 2006) and its therapeutic potential has been certified by its antipyretic (Owoyele and Adeyeye, 2015), analgesic, anti-inflammatory (Owoyele *et al.*,

2014) and neuroprotective effects (Abdulmajeed *et al.*, 2016). Studies have shown that different types of honey (Manuka and Tualang types of honey) possess anti-ulcer effects. However, no such report is available for Unilorin honey, a multiflora honey gotten from nectars of local plants (Abdulrahaman *et al.*, 2013). Therefore, the index study investigated gastro-protective effects of Unilorin honey in male Wistar rats.

Materials and Methods

Honey was obtained from Unilorin apiary, University of Ilorin, Nigeria.

Drug and Chemicals

Indomethacin and Cimetidine were procured from Fiolu Pharmaceutical Store, Gambari Road, Ilorin, Nigeria while other chemicals used in the index study were products from Sigma-Aldrich CHEME GmbH, Steinheim, Germany.

Laboratory animals

Forty Male Wistar rats, with weight range of 190-230 g were obtained from the Animal Holding Unit of the Department of Biochemistry, University of Ilorin. The rats were acclimatized for two weeks. They were fed on rat pellet (Premier feed Ltd, Ibadan) and water *ad libitum*. They were subjected to standard laboratory conditions and natural photoperiod of 12h light; dark cycle: temperature: 28-31oC: humidity: 50-55%. The NIH Publication No. 85-23 guidelines (NIH Publication revised, 1985) was used in the experimental protocol and animal handling.

Preparation and administration of Indomethacin

Indomethacin capsules (100 mg) was dissolved in normal saline (10 ml) and given by oral gavage at 20 mg/kg body weight.

Animal Grouping and Administration of the honey

Two models were adopted for ulcer induction, the HCl/ethanol and the Indomethacin models. There

were 20 rats in each model. This was further divided into 4 groups, each made of 5 rats. For each model, one group (D) had a high dose of honey at 1000mg/kg, another group (C) had a low dose of honey administered at 250mg/kg, while the third group (B-the reference group) was given Cimetidine at a dose of 100 mg/kg. The last group (A-control) was given 0.5 ml of normal saline.

HCl/ethanol model (1.2ml of 0.15NHCl in 70% ethanol) Group A- (control) normal received 0.5ml of normal saline Group B- received 100 mg/kg of Cimetidine Group C- received an oral administration of 250 mg/kg b.w. of Unilorin honey (UH) Group D - received an oral administration of 1000 mg/kg b.w. of Unilorin honey (UH). Indomethacin model Group A- (control) normal and received 0.5ml of normal saline Group B- received 100 mg/kg of Cimetidine Group C- received an oral administration of 250 mg/kg b.w. of Unilorin honey Group D - received an oral administration of 1000 mg/kg b.w.

These administrations were done over a period of 12days as a pre-treatment before ulcer induction. Then, the rats were fasted for 24hours with access to water only. Ulcer induction was achieved by the administration of 1.2ml of 0.15N HCl in 70% ethanol for the HCl/Ethanol group, and 20 mg/kg of indomethacin for the indomethacin group.

The administered drug and Unilorin honey were suspended in vehicle (normal saline) solution and administered orally by gastric gavage once in a day for 12 days

HCl/ethanol model (1.2ml of 0.15NHCl in 70% ethanol)

Group A- (control) normal and received 0.5ml of normal saline

Group B- received 100 mg/kg of Cimetidine

Group C- received an oral administration of 250 mg/kg b.w. of Unilorin honey (UH)

Group D - received an oral administration of 1000 mg/kg b.w. of Unilorin honey (UH)

Indomethacin model

Group A- (control) normal and received 0.5ml of normal saline

Group B- received 100 mg/kg of Cimetidine

Group C- received an oral administration of 250 mg/kg b.w. of Unilorin honey

Group D - received an oral administration of 1000 mg/kg b.w. of Unilorin honey

The drug and Unilorin honey were suspended in vehicle (normal saline) solution and were administered orally by gastric gavage once daily for 12 days.

Grading of ulcer

The method described by Berry et al. (1988) was used in grading the ulcer: The rats were sacrificed using ether. The abdomen was dissected and the stomach removed and cut open along the greater curvature. The stomach was then washed clean using normal saline, spread on a plain white material and examined for ulceration. The ulcer index, which is an expression of the severity of gastric mucosa lesion was determined and graded as follows:

- 0 – no lesion
- 0.5 – hyperaemia
- 1.0 – slight lesion
- 2.0 – severe lesion
- 3.0 – very severe lesion
- 4.0 – mucosa full of lesion

Percentage inhibition was calculated as

Percentage inhibition=

$$\frac{\text{mean ulcer index (Control)} - \text{mean ulcer index (Treated)}}{\text{mean ulcer index (Control)}} \times 100$$

Determination of Secondary Metabolites

Unilorin honey was subjected to standard chemical test as described for Tannins, Phlobatannins, Steroids and Cardiac glycosides (Trease and Evans, 1989), Saponins (Sofowora, 1993), Flavonoids (Sofowora, 1993; Harbone, 1998) and Terpenoids (Sofowora, 1993).

Measurement of gastric acidity

Isolation of stomach and collection of gastric juice

At the end of experimental period, the animals were anaesthetized using over dose of ether the abdominal region were dissected and the stomachs of the animals were collected and cut open along the greater curvature. The gastric content was collected, centrifuged for 10 minutes at a speed of 200m/s. A quantifiable volume (1ml) of supernatant from each rat was analyzed for hydrogen ion concentration by titrating it against 0.01M of NaOH to a pH 7.0 which was detected by using phenolphthalein as indicator (2 drops). When titrated it gave a pinkish colour. The acid content was expressed in Meq/L (Shay *et al.*, 1945) using the formula below:

$$CaVa = CbVb$$

Where; Ca=Concentration of Acid
Cb= Concentration of Base
Va= Volume of Acid
Vb= Volume of Base

Histological study

The gastric tissues were fixed in 10% formalin. The tissues were processed using a tissue processor and the processed tissues were embedded in paraffin block and little thick sections were cut using a microtome (Yidi-Jinhua Zhejiang, China). These sections were stained using hematoxylin and eosin stain. The slides were then examined under the microscope for morphological changes such as inflammation, congestion, edema, haemorrhage and erosion or positive results such as regeneration and protection of mucosal layer and their photomicrographs were taken.

Statistical analysis

The results are expressed as Mean ± SEM. Data were subjected to analysis of variance (ANOVA) followed by Waller-Duncan *post hoc* test using SPSS v 16 statistical package. p<0.05 was considered statistically significant.

Results

The result of the secondary metabolite constituents of University of Ilorin honey (Table 1) shows that Saponins, Flavonoids, Steroids, Terpenoids and Cardiac glycosides were detected while Tannins, Phlobatannins were not detected. Figure 1 shows the effect of Unilorin honey on gastric ulceration in HCL/Ethanol ulcer model. The control group (pre-treatment with normal saline) had a high ulcer index of 6.80 ± 0.20. Pre-treatment of the animals with different doses of University of Ilorin honey (250 mg/kg b.w and 1000 mg/kg b.w) showed a significant dose dependent reduction in ulcer index of 4.60 ± 0.24 (250 mg/kg b.w.) and 2.4 ± 0.24 (1000 mg/kg b.w.) with a percentage ulcer inhibition of 64.7% (1000 mg/kg b.w) and 32.9% (250mg/kg b.w). The group pre-treated with 1000 mg/kg b w compared favourably with the group that was pre-treated with the reference drug (Cimetidine) (2.4 ± 0.24 Vs 2.0 ± 0.45)

Figure 2 shows the effect of Unilorin honey (UH) on gastric ulceration in Indomethacin induced ulcer model. The control group that was pre-treated with normal saline had a mean ulcer score of 7.2±0.37. Pre-treatment of animals with different doses of Unilorin honey (250mg/kg bw and 1000mg/kg bw) caused a significant (p<0.05) dose dependent reduction in ulcer formation induced by Indomethacin (5.00 ± 0.32 and 3.20 ± 0.37 respectively) with percentage inhibitions of 30.6% and 55.6% respectively. The group pre-treated with 1000 mg/kg b w of Unilorin honey (UH) compared favourably with Cimetidine pre-treated group with a percentage ulcer inhibition of 69.4% and a mean ulcer score of 2.20 ± 0.24.

In Table 2 and 3, there is no significant reduction in gastric acid secretion between the control and other treated groups in both models of ulcer induction (HCl/Ethanol and Indomethacin models).

Figure 3 and 4 showed the histological preparation of the stomach from both HCl/Ethanol and Indomethacin models respectively. The normal saline pre-treated group showed damage in the gastric mucosa with many ulcerated areas while the histology of the gastric mucosa of the groups treated with 250 mg/kg b.w, 1000 mg/kg b.w. of Unilorin honey and the cimetidine showed a well preserved mucosa with reduced ulcer areas.

Table 1: Secondary Metabolite constituents of Unilorin honey

Secondary Metabolites	Observation
Saponins	+
Flavonoids	+
Steroids	+
Terpenoids	+
Cardiac glycosides	+
Tannins	-
Phlobatannins	-

+ = Present; - = Absent

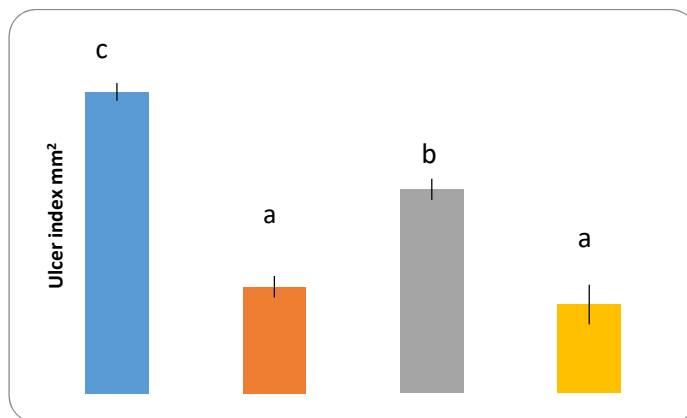


Fig 1: Effect of Unilorin honey (UH) on Ulcer index in HCl/Ethanol induced ulcer.

A is the control; B is cimetidine treated group; C is UH 250 mg/kg treated group; D is UH 1000 mg/kg treated group. Values are means of five determinations ± SEM. Bars with different letters are significantly different at p<0.05)

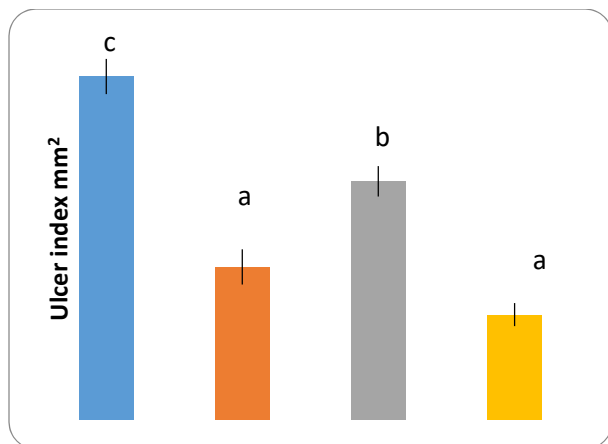


Figure 2: Effect of Unilorin honey (UH) on ulcer index in Indomethacin induced ulcer.

A is the control; B is cimetidine treated group; C is UH 250 mg/kg treated group; D is UH 1000 mg/kg treated group. Values are means of five determinations ± SEM. Bars with different letters are significantly different at p<0.05)

Table 2: Effect of Unilorin honey (UH) on gastric acidity in HCl/Ethanol induced ulcer

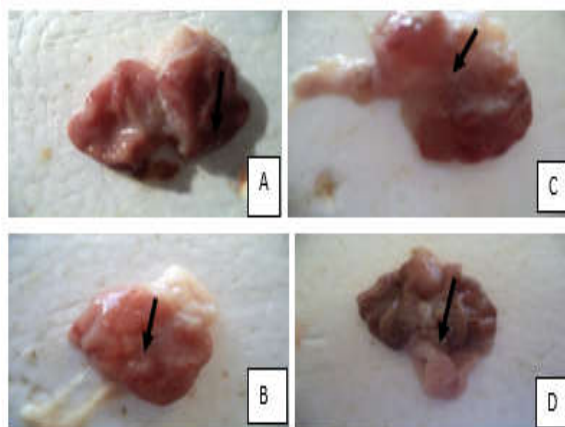
Groups (N)	SG	Dosage	N	Gastric acidity (X 10 ⁻³ Meq/L)
A	Normal saline	0.5 ml	5	1.70±0.36 ^{ab}
B	Unilorin honey	1000 mg/kg	5	2.22±0.28 ^b
C	Unilorin honey	250 mg/kg	5	1.66±0.98 ^{ab}
D	Cimetidine	100 mg/kg	5	1.31±0.11 ^a

N= groups; A is the control; B is Unilorin Honey (1000 mg/kg) pre-treated; C is Unilorin honey (250 mg/kg) pre-treated and D is cimetidine (100 mg/kg) pre-treated. n= number of rats, SG= substance given. Values are means of 5 determinations ± SEM. Values with the same superscript in the same column are not significantly different (p<0.05)

Table 3: Effect of Unilorin honey (UH) on gastric acidity in Indomethacin induced ulcer

Groups (N)	SG	Dosage	N	Gastric acidity (X 10 ⁻³ Meq/L)
A	Normal saline	0.5 ml	5	1.83±0.39 ^a
B	Unilorin honey	1000 mg/kg	5	1.56±0.15 ^a
C	Unilorin honey	250 mg/kg	5	1.74±0.35 ^a
D	Cimetidine	100 mg/kg	5	1.27±0.94 ^a

N= groups; A is the control; B is University of Ilorin Honey (1000 mg/kg) pre-treated; C is University of Ilorin honey (250 mg/kg) pre-treated and D is cimetidine (100 mg/kg) pre-treated. n= number of rats, SG= substance given. Values are means of 5 determinations ± SEM. Values with the same superscript in the same column are not significantly different (p<0.05)



Photograph 1: Stomach of Rats pre-treated with normal saline, Unilorin honey and cimetidine in HCl/Ethanol model. A: control, mucosal full of ulcer; B: Unilorin honey (1000 mg/kg), smooth mucosal; C: Unilorin honey (250 mg/kg), mucosal with few ulcers; D: Cimetidine (100 mg/kg) pretreated group, mucosal with few ulcers.

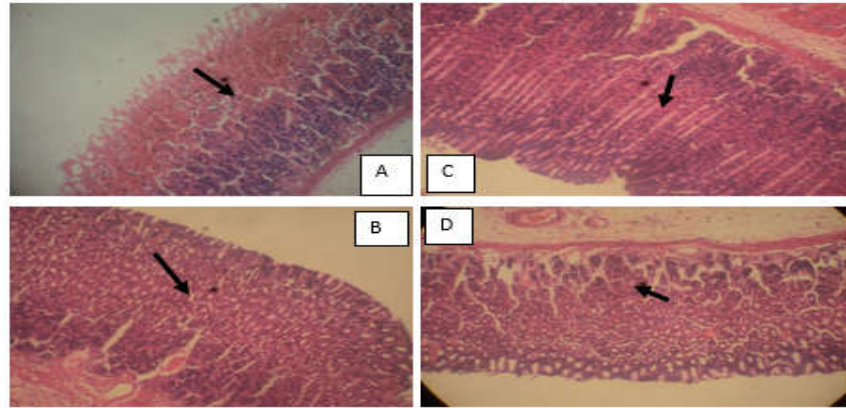
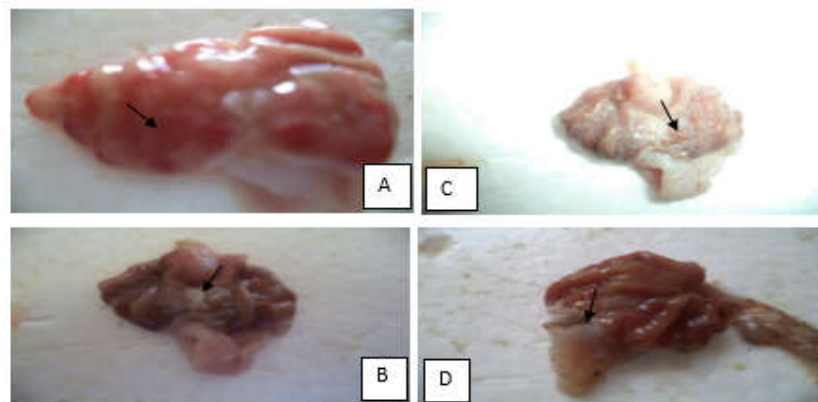


Fig 3: Photomicrograph of section of stomach of rats pre-treated with normal saline, University of Ilorin honey and cimetidine in HCl/ Ethanol model. A: control, ulcerated mucosal; B: Unilorin honey (1000 mg/kg), smooth mucosal; C: Unilorin honey (250 mg/kg), mucosal with few ulcers; D: Cimetidine (100 mg/kg) group, mucosal with few ulcers. Magnification X 100



Photograph 2: Stomach of Rats pre-treated with normal saline, Unilorin honey and cimetidine in indomethacin model. A: control, mucosal with visible ulcers; B: University of Ilorin honey (1000 mg/kg), mucosal with no traceable visible ulcers; C: Unilorin honey (250 mg/kg), mucosal with few ulcers; D: Cimetidine (100 mg/kg) pre-treated group, mucosal with few ulcers.

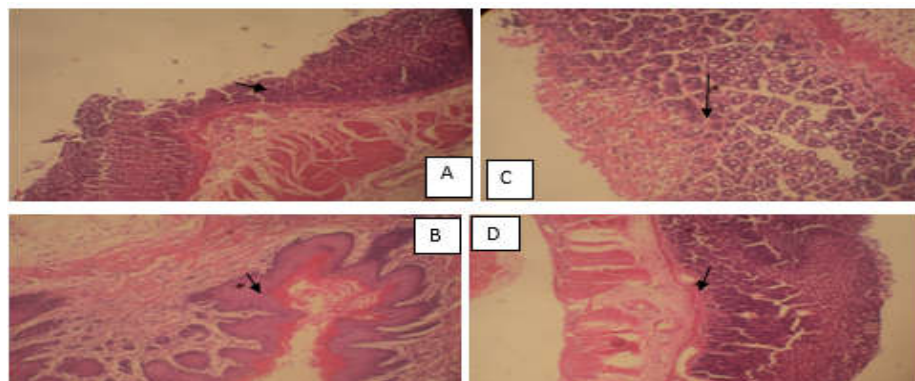


Fig 4: Photomicrograph of section of stomach of rats pre-treated with normal saline, University of Ilorin honey and cimetidine in indomethacin model. A: control, ulceration extensively covering large area of the mucosal; B: Unilorin honey (1000 mg/kg), no ulceration, there is keratinized squamous epithelium; C: Unilorin honey (250 mg/kg), mucosal with fewer ulcerations, ulcers are focal ulceration, there is presence of squamous metaplasia; D: Cimetidine (100 mg/kg) pre-treated group, mucosal with few ulcers and the presence of hyperplasia. Magnification X 100

Discussion

Several drug-induced ulcer models have been used to investigate gastro-protective potentials of medicinal agents, these models include NSAID induced gastric ulcer, ethanol-induced gastric ulcers, HCl-induced gastric ulcer, histamin-induced gastric ulcers etc (Hay *et al.*, 1942; Djahanguiri, 1969; Levine, 1971; Oates and Hakkinen, 1988). Indomethacin is a non-steroidal anti-inflammatory drug (NSAID). Indomethacin ulcer induced model used in this study is one of the most acceptable models used to study ulceration in the gastrointestinal tracts of animals it was also used in this study because it is the second most common cause of ulcer after *Helicobacter pylori*.

The underlying mechanisms of indomethacin-induced ulcer involve inhibition of prostaglandin synthesis (Allen *et al.*, 1993; Adinortey *et al.*, 2013), generation of reactive oxygen species (ROS) in response to oxidative stress (Saini *et al.*, 2012) etc. Prostaglandin plays a protective role by stimulating bicarbonate and mucus secretion, repair, regulate mucosal cell turnover and maintain mucosal blood flow. Inhibition of prostaglandin synthesis by indomethacin could lead to increased susceptibility of gastric mucosa to injury, thereby causing gastric ulceration (Adinortey *et al.*, 2013).

The HCl/Ethanol model is also one of the method used to study ulceration in gastric mucosa. This model induces ulcer by causing increased vascular permeability which in turn causes the release of vasoactive substances like histamin, pepsin (Mahmood *et al.*, 2011). This model also mediate its ulcerogenic effects by decreasing bicarbonate and mucus production and increasing production of reactive oxygen species.

In the present study, the significant increase in mean ulcer score in normal saline pre-treated group shows that indomethacin and HCl/Ethanol cause gastric ulceration, but pre-treatment of the

rats with different doses of University of Ilorin honey reduced the susceptibility of gastric mucosa to injury caused by indomethacin and HCl/Ethanol. The observed decrease in mean ulcer index in Unilorin honey pretreated groups in both indomethacin and HCl/Ethanol models in a manner similar to that of cimetidine group suggests that Unilorin honey has gastro-protective effect which is in line with previous study that documented anti-ulcer effect of honey (Nasutia *et al.*, 2006).

The gastroprotective effect of this honey may not be unconnected to the presence of some secondary metabolites which include Flavonoids, Terpenoids, Steroids, Saponinns and Cardiac glycosides. This finding was supported by other studies that stated that biological actions of honey are attributed to their polyphenolic compounds (Alvarez-suarez *et al.*, 2013). Polyphenolic compounds have been reported to have gastroprotective effects probably via stimulation of prostaglandin E₂ formation (Alanko *et al.*, 1999), and/or through their ability to scavenge free radicals or stimulating antioxidant enzymes activities (Di Carlo *et al.*, 1999). The presence of these secondary metabolites in University of Ilorin type of honey may among other factors be responsible for its anti-ulcer effect.

Anti-ulcer effect of Unilorin honey may also be linked to its physicochemical properties like carbohydrate, ash, protein which has earlier been reported (Ekanem *et al.*, 2006; Okeola *et al.*, 2015). Its high sugar (carbohydrate) content in comparison with other constituents (Ekanem *et al.*, 2006; Okeola *et al.*, 2015) may offer some protection on gastric mucosal because previous study has reported that carbohydrate causes reduction in acid-back diffusion and lipid peroxidation in the gastric mucosa (Hung and Neu, 1997).

The ash content of honey represents their mineral and trace element constituents (Buba *et al.*, 2013). Previous studies (Ekanem *et al.*, 2006; Okeola *et al.*

al., 2015) have reported that University of Ilorin honey has ash content that is in agreement with standards of Codex Alimentarius. Some of these minerals and trace elements might serve as antioxidants and reports from researches have documented that antioxidants protect gastric mucosal and inhibit progression of gastric ulcer (Dekkers *et al.*, 1996).

The mechanism for the anti-ulcer effect doesn't seem to include inhibition of acid secretion because the result from the study revealed no significant difference in gastric acid secretion between the normal saline-pretreated group and Unilorin honey pre-treated groups. Therefore, the gastroprotective effect of Unilorin honey recorded in this study could be via activation of prostaglandin synthase through the cyclooxygenase pathway (Rainford, 1987), which in turn stimulates the secretion of bicarbonate and mucus, or it might have acted by causing decrease in pepsin or reactive oxygen species production, hence, decreased susceptibility of the mucosa to injury. Further studies will be carried out to elucidate mechanisms used by this type of honey in preventing ulcer

Conclusions

This study concludes that Unilorin honey has gastroprotective effect which is not due to alteration in gastric acidity.

Acknowledgement

We acknowledge the efforts of Mrs Olawale-Bello for providing technical assistance in the course of this research work

Competing interests

The authors declare no conflict of interest. This study was funded by the authors. No financial assistance was gotten from any funding agency or Institution.

References

- Abdulmajeed, W.I., Suleiman, H.B., Zubayr, M.O., Imam, A., Amin, A., and Abayomi, S. (2016). Honey prevents neurobehavioural deficit and oxidative stress induced by lead acetate exposure in male Wistar rats- a preliminary study. *Metabolic Brain Disease*, 31, 37-44
- Abdulrahman, A.A., Solomon, O.R., Adeyemi, S.B., Liadi, M.T., Ahmed, R.N., Belewu, M.A., and Oladele, F.A. (2013). Melisopolynological analysis of honey samples from *jatropha* plantation and Unilorin apiary farm. *International Journal of Phytofuels and Allied Sciences*, 2(1), 81-92
- Adinortey, M.B., Ansah, C., Galyuon, I. and Nyarko, A. (2013). *In vivo* models used for evaluation of potential antigastroduodenal ulcer agents. *Ulcer*, 2013 article ID 796405, 12 pages
- Akharaiyi, F.C. and Lawal, H.A. (2016). Physicochemical Analysis and mineral contents of honey from farmers in Western States of Nigeria. *Journal of Natural Sciences Research*, 6(19), 78-84
- Alanko, J., Riutta, A., Holm, P., Mucha, I., Vapata, H. and Metsa-Ketala, T. (1999). Modulation of arachidonic acid metabolism by phenols: Relation to their structure and antioxidant/pro-oxidant properties. *Free Radic Biol Med*, 26, 193-201
- Allen, A., Flemstron, G., Garner, A. and Kivilaakso, E. (1993). Gastroduodenal mucosal protection. *Physiol Lev*, 73, 823-857
- Alvarez-suarez, J.M, Giampieri, F and Battino, M. (2013). Honey as a source of dietary antioxidants, structures, bioavailability and evidence of protective effects against

human chronic diseases. *Curr Med Chem*, 20 (5), 621-638

Alzubier, A.A. and Okechukwu, P.N. (2011). Investigation of anti-inflammatory, antipyretic and analgesic effects of Yemeni sidr honey. *World Acad Sci Engr Technol*, 80, 47-52

Amandeep, K., Robin, S., Ramica, S. and Sunil, K. (2012). Peptic Ulcer: A Review on Etiology and Pathogenesis. *International Research Journal of Pharmac.*, 3(6), 34-38.

Azim, M.K., Simjee, S.U., Perveen, H. and Mesaik, M.A. (2007). Anti-nociceptive activity of natural honey in thermal-nociception models in mice. *Phytother Res*, 21(2), 194-197.

Berry, C.N., Prouteau, M. and Lloyd, K.G. (1988). Sulphasalazine and PhCL28A inhibit the formation of ethanol and phenylbutazone induced gastric ulcer lack of involvement of endogenous prostaglandin. *Br. J. Pharmacol*, 93, 465-471.

Biglari, B., Moghaddain, A., Santos, K., Blaser, G., Buchler, A., Jansen, G., Langler, A., *et al.* (2013). Multicentre perspective observational study on professional wound care using honey (medi honey™). *Int Wound J*, 10(3), 252-259

Bogdanov, S., Ruoff, K. and Persano-oddo, L. (2004). Physicochemical methods for the characterization of unifloral honeys: A review. *In Apidologie*, 35(1), S4-S17

Buba, F., Gidado, A. and Shugaba, A. (2013). Analysis of biochemical composition of honey samples from North-East Nigeria. *Biochem Anal Biochem*, 2(3), 1-7

Dekkers, J.C., Van Doomen, L.J., Han, C. and Kemper, G. (1996). The role of antioxidant, vitamins as enzymes in the prevention of exercise-induced muscle damage. *Sport Medicine*, 21, 213-238

Di Carlo, G., Mascolo, N., Izzo, A.A. and Capasso F. (1999). Flavonoids: Old and New aspects of a class of natural therapeutic drugs. *Life Sci*, 64, 337-353

Djahanguiri, B. (1967). The production of acute gastric ulceration by indomethacin in the rat, *Scandinavian Journal of Gastroenterology*, 4(3), 265-267

Ekanem, J.T., Majolagbe, O.R., Sulaiman, F. A. and Muhammad, N.O. (2006). Effects of honey-supplemented diet on the parasitemia and some enzymes of *Trypanosoma brucei*-infected rats. *African Journal of Biotechnology*, 5(17), 1557-1561

Goyal, R.K. (2008). *Element of Pharmacology*, B.S. Shah Prakashan, Ahmedabad, 7th Edition

Gunduz, A., Eraydin, I., Turkmen, S., Kalkan, O.F., Turedi, S., Eryigit, U. and Ayar, A. (2013). Analgesic effects of mad honey (grayanotoxin) in mice models of acute pain and painful diabetic neuropathy. *Hum Exp Toxicol*, 33(2), 130-135

Harborne, A.J. (1998). *Phytochemical methods: A guide to modern techniques of plant analysis*. Chapman & Hall, London, 279

Hay, L.J., Varco, R.L., Code, C.E. and Wangenstein, O.E. (1942). Experimental production of gastric and duodenal ulcers in laboratory animals by intramuscular injection of histamine in beeswax. *The Journal of Surgery, Gynaecology and Obstetrics*, 74, 70-182

- Hung, C.R. and Neu, S.L. (1997). Acid-induced gastric damage in rats is aggravated by starvation and prevention by several nutrients. *Journal of Nutrition*, 127(4), 630-636
- Kassim, M., Mansor, M., Al-Abd, N. and Mohd Yusoff, K. (2012). Gelam Honey Has a Protective Effect against Lipopolysaccharide (LPS)- Induced Organ Failure; *Int J Mol Sci*, 13(5), 6370–6381.
- Kassim, M., Achoui, M., Mustafa, M.R., Monsur, M. and Yusoff, K.M. (2010). Inhibitory effects of Gelam honey and its extracts on nitric oxide and prostaglandin E₂ in inflammatory tissues. *Fitoterapia*, 81(8), 1196-1201
- Khazael, M. and Salehi, H. (2006). Protective effects of *Fulcaria vulgaris* extract on ethanol induced gastric ulcer in rats. *Iranian Journal of Pharmacology & therapeutics IJPT*, 5,43-46
- Levine, R.J. (1971). A method for rapid production of stress ulcers in rats, in *Peptic Ulcer*, C.J. Pfeiffer, Ed.: Munksgaard, Copenhagen, Denmark, 92-97
- Lozano, R. (2010). Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study. *Lancet*, 380(9859), 2012; 2095–128.
- Mahmood, A.A., Al-Bayat, F.H., Salmah, T., Nor Syuhada, A.B., Harita, H. and Mughrabi, F. (2011). F₃ enhancement of gastric ulcer by *areca catechu* nut in ethanol induced gastric mucosa injuries in rats. *Journal of Medicinal Plant Research*, 5(12), 2562-2569
- Malferteiner, P., Chan, F.K. and McColl, K. E. (2009). Peptic ulcer disease. *The Lancet*, 374, 1449-1461
- Molan, P.C. (2001). Why honey is effective as a medicine-2. The scientific explanation of its effects. *Bee World*, 82(1), 22-40
- Nasutia, C., Gabbianellib, R., Falcionib, G., Cantalamessa, F. (2006). Antioxidative and gastroprotective activities of anti-inflammatory formulations derived from chestnut honey in rats. *Nut Res*, 26, 130-137
- Oates, P.J. and Hakkinen, J.P. (1988). Studies on the mechanisms of ethanol-induced gastric damage in rats. *Gastroenterology*, 94(1), 10-21.
- Okeola, F.O., Afolabi, B.T., Liadi, M.T. and Apo, G.S. (2015). Comparative study of Physico-Chemical properties of pure honey harvested from *Jatropha curcas* plantation and honey fortified with Moringa and Ginger. *International Journal of Phytofuels and Allied Sciences*, 4(1), 81-92
- Omoya, F.O. and Akharaiyi, F.C. (2010). A pasture honey trial for antibacterial potency on some selected pathogenic bacteria. *Journal of Natural Products*, 3, 05-11
- Owoyele, B.V. and Adeyeye, E.O. (2015). Antipyretic effect of honey in Albino rats. *Utilization and Management of Medicinal Plants*, 3, 315-320
- Owoyele, B.V., Oladejo, R.O., Ajomole, K., Ahmed, R.O. and Mustapha, A. (2014). Analgesic and anti-inflammatory effects of honey: the involvement of autonomic receptors. *Metab Brain Dis*, 29, 167-173.
- Prabha, T., Dorababu, M., Goel, S., Agarwal, P.K., Singh, A., Joshi, V.K. and Goel, R.K. (2009).

Effect of methanolic extract of *Pongamia pinnata* linseed on gastro-duodenal ulceration and mucosal offensive and defensive factors in rats. *Indian J. Exp. Biol*, 47, 649-659

Page | 4125

Rainford, K.D. (1987). The effects of 5-lipoxygenase inhibitors and leukotriene antagonists on the development of gastric lesions induced by nonsteroidal anti-inflammatory drugs in mice". *Agents and Actions*, 2(3-4), 316-319

Saini, A., Sharma, N. and Sharma, R. (2012). Animal models to evaluate the cause behind gastric ulcers. *IRJP*, 3(8), 34-36

Sharifi-Rad, M., Fokou, P.V.T., Sharopov, F., Martorell, M., Ademiluyi A.O., Rajkovic, J, Salehi, B., Martins, N., Iriti, M., Sharifi-Rad, J. (2018). Antiulcer agents: from plant extracts to phytochemicals in healing promotion. *Molecules*, 23, 1751.

Shay, H., Komarov, S.A., Fels, S.S., Meranze, D., Gruenstein, M. and Siptet, M. (1945). *Gastroenterology*, 5, 43-61.

Sofowora, A. (1993). Medicinal plants and traditional medicines in Africa. 2nd edn. Spectrum Book Limited Ibadan, Nigeria, 134-156

Sung, J.J., Kuipers, E.J., El-Serag, H.B. (2009). Systematic review: the global incidence and prevalence of peptic ulcer disease. *Alimentary Pharmacology and Therapeutics*, 29(9), 938-946

Trease, G.E. and Evans, W.C. (1989). A textbook of Pharmacognosy, 13th edn. Bailliere-Tindall Ltd., London