Antiulcer effect of Acacia nilotica seedpod aqueous extract on experimentally induced ulcer
Azeemat Titilola Abdulazeez and Elizabeth Abidemi Balogun
1Department of Biological Sciences, Al-Hikmah University, Ilorin, Nigeria. E-mail: azeemahtitilola@alhikmah.edu.ng
2Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. E-mail: ea_balogun@yahoo.com

Abstract
Acacia nilotica is a plant used for the treatment of various ailments. This research was conducted to assess the anti-ulcerogenic property of A. nilotica seedpod using ethanol and indomethacin as ulcer-inducing agents. Experimental rats were grouped and orally pre-treated with varying doses of aqueous extract of A. nilotica seedpod and ranitidine (10 mg/kg bw) before the ulcer was induced using ethanol. Another set of rats were also post-treated with graded doses of aqueous extract of A. nilotica seedpod and ranitidine following induction of ulcer using indomethacin. The effect of the extract on ulcer indices, inflammatory markers, antioxidant defense systems was monitored. Aqueous extract of A. nilotica seedpod gave optimum gastroprotection (42.86%) and ulcer healing (70.25%) effect at 50 mg/kg bw. LD_{50} of the extract was greater than 3,000 mg/kg bw and secondary metabolites like phenolics, glycosides are present in the aqueous extract. The findings of the study show that A. nilotica seedpod has anti-ulcerogenic activity, thus, supporting its folkloric use for the treatment of peptic ulcer disease (PUD).

Keywords: A. nilotica, Ethanol, Indomethacin, Anti-ulcerogenic, Phytochemicals

1. Introduction
Disorders affecting the gastrointestinal tract and the accessory organs of digestion are collectively known as gastrointestinal diseases (Reed and Wickham, 2009) and these affect about 10% of the entire world (Day, 2019). Peptic injury that leads to mucosal lesion in the stomach or proximal duodenum is known as Peptic Ulcer Disease (PUD) (Lanas and Chan, 2017). PUD has been shown to occur due to a negative balance between the factors that protect the gastric mucosal and factors that could damage it (Sverden et al., 2019). Damaging factors include Helicobacter pylori, alcohol and non-steroidal anti-inflammatory drugs (Narayanan et al., 2018). Nwokediuko et al. (2012) showed that there is a decrease in the occurrence of duodenal ulcers in contrast to an increase in gastric ulcers in a study covering 15 years between 1995 and 2010 in Nigeria.
Several therapeutic options have been employed for the treatment and management of PUD (Brunton, 2001). Antacids, selective antagonists of gastrin receptors, histamine-H₂ receptors, Proton Pump Inhibitors (PPIs), mucoprotective medicines, eradication of Helicobacter pylori, and analogues of prostaglandins are some of the options used (Daley, 2012).

Peptic ulcer therapy through combination of drugs has not been very effective due to non-compliance of drug regimen by patients and possible side effects of the drugs due to their long term use (Peura, 2004). Treatment of gastric disorders with medicinal plants is quite common in traditional medicine worldwide (Sachan et al., 2018).

Plants produce a large number of secondary metabolites that are vital for animal and human health, as well as model chemicals for drug synthesis or intermediates for synthetic pharmaceuticals (Atanasov et al., 2015).

Acacia nilotica (L.) is a popular plant that grows naturally; it is used in both crop and animal farming systems (Shittu, 2010). Many scientific studies have been used to ascertain its various pharmacological claims; these include anti-diabetic, hypolipidemic, antileishmanial effects (Rather and Mohammad, 2015).

A. nilotica Seedpod is boiled as a decoction and taken with or without milk to treat a peptic ulcer by Traditional Medicine Practitioners in Nigeria. Therefore, this study was conducted to ascertain this claim by investigating both its protective and healing effects on PUD.

2. Materials and methods

2.1. Plant materials and animals

A. nilotica seedpods were obtained from a local market in Kaduna, Nigeria. They were identified and authenticated at the Department of Plant Biology, University of Ilorin, Nigeria where a voucher specimen (UILH/002/1174) was prepared and deposited in the Herbarium. The seedpods were pulverized into a fine powder and 150 g was macerated in 1500 mL of distilled water for 24 h. The filtrate obtained was evaporated to dryness to obtain a semi solid residue.

2.2. Laboratory animals

Wistar rats (Rattus norvegicus) of both sexes weighing 180.6 ± 20g were procured from the Animal holdings, Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. They were acclimatized for seven days before the commencement of the study.

2.3. Ethical approval

An approval number UERC/ASN/2015/118 was issued by the University of Ilorin Ethical Clearance Committee for animal use. They were maintained under standard laboratory conditions with free access to standard animal feeds (Top Feed, Nigeria) and clean water.

2.4. Screening of secondary metabolites in A. nilotica seedpod

The qualitative and quantitative analysis of aqueous extract A. nilotica seedpod was determined using standard procedures.

2.5. Acute toxicity

LD₅₀ was determined using the limit test dose (OECD, 2001). A aqueous extract of A. nilotica seedpod (3000 mg/kg bw) was administered orally to five rats, one rat at a time. The animals were observed for 14 days. LD₅₀ was expressed based on the survival rate of the animals.

2.6. Induction of ulcer, animal groupings, and extract administration

Thirty-six rats of both sexes were used; they were acclimatized for seven days and were fasted for 18 hours before the start of the study. Thirty (30) rats out of the thirty-six (36) received 10 mg/ kg body weight (bw) of indomethacin for three days to induce ulcer while the remaining six rats were used as control. Indomethacin-induced ulcerated rats were randomly divided into five groups of six rats each as follows: Group 1 (Control group) received distilled water; Group 2 (Ulcerated not treated) received distilled water; Group 3 (Ulcerated) was administered 10 mg/ kg bw ranitidine; Group 4 (Ulcerated) was administered 12.5 mg/ kg bw of aqueous
extract of A. nilotica pod; Group 5 (Ulcerated) was administered 25 mg/kg bw of aqueous extract of A. nilotica pod and Group 6 (Ulcerated) was administered 50 mg/kg bw of aqueous extract of A. nilotica pod. The animals were treated accordingly for seven days; fasted overnight after the last treatment to ensure complete gastric emptying. The animals were sacrificed under diethyl ether anesthesia.

2.7. Gastroprotective study of aqueous extract of A. nilotica pod

Another thirty-six (36) animals of both sexes were fasted for 24 h and randomly divided into six groups of six rats each and were given treatment for seven days as follows: Group 1 (Control group) received distilled water; Group 2 was administered distilled water while Groups 3, 4, 5, and 6 were administered 10 mg/kg bw of ranitidine, 12.5, 25, and 50 mg/kg bw of aqueous extract of A. nilotica pod respectively for seven days and one hour after the last treatment, all animals in groups 2-6 were induced with 1 mL of 95% ethanol orally (Morimoto et al., 1991). The animals were sacrificed sixty minutes after induction under diethyl ether anesthesia.

Rat abdomens were cut open, the esophagus and pyloric were clamped before the stomach was removed. A cut was made at the pyloric; the gastric content was poured into a centrifuge tube and the stomach was washed with 3 mL normal saline. This was centrifuged at 1,000 rpm for 10 min; the supernatant was used for gastric secretion evaluation. A portion of the stomach was weighed, rinsed and homogenized using buffered saline (pH 7.4), this was then centrifuged at 2,500 rpm for 10 min, and the supernatant was used for biochemical assays.

2.8. Evaluation of gastric acid secretory indices

Volume and pH of gastric content supernatant were noted as the gastric juice volume and gastric pH respectively (Da Silva et al, 2015). One milliliter of the supernatant was then pipetted into a conical flask; few drops of methyl orange indicator were added and then titrated against 0.01 N NaOH until there was a color change to yellow, the volume of alkali used was reported as free acidity. Few drops of phenolphthalein solution were then added and titration continued until a definite red tinge color appeared. The volume of alkali used corresponds with the total acidity (Srivastava et al., 2010). Acidity was calculated using the formula:

\[
Acidity (\text{Meq/L/100 g}) = \frac{\text{Volume of NaOH} \times \text{Normality of NaOH}}{0.1} \times 100
\]

2.9. Pepsin activity of the gastric juice was estimated as described by Anson (1938)

2.9.1. Gastric wall studies

Ulcer index was calculated using the method described by Tanaka et al. (1993). Percentage of ulcer healing/ulcer inhibition was also estimated (Sánchez-Mendoza et al., 2011). Adherent Mucin concentration was estimated as described by Corne et al. (1974).

2.9.2. Determination of biochemical parameters

Myeloperoxidase activity in the gastric mucosa was determined using the procedure described by Weiss et al. (1982). Superoxide Dismutase Activity (Misra and Fridovich, 1972); Catalase Activity (Beers and Sizer, 1952); Glutathione peroxidase (GPx) activity (Rotruck et al., 1973); glutathione transferase (GST) activity (Habig et al., 1974), the concentration of reduced glutathione concentration (GSH) was determined as described by Ellman (1959). The rate of lipid peroxidation was assessed by quantifying malondialdehyde levels as described by Reilly and Aust (2001).

2.9.3. Statistical analysis

Data were expressed as a mean of six replicates ± SEM and were analyzed using one-way ANOVA followed by Tukey multiple comparisons test using GraphPad statistical package version 6.0. Values were considered significant at 95% confidence interval (p < 0.05).

3. Results

3.1. Preliminary evaluation of secondary metabolites

Table 1 shows the secondary metabolites present in A. nilotica seedpod aqueous extract. Alkaloids, saponins, tannins, phenolics, flavonoids, and glycosides were detected while terpenoids, anthraquinones, and volatile
oils were not detected. Total phenol and flavonoid contents are 110.0 mg/g gallic acid and 16.24 mg/g quercetin equivalent respectively while saponin concentration was 6.82 mg/ml.

| Table 1: Secondary metabolites present in aqueous extract of A. nilotica seedpod |
|-----------------------------------|------------------|
| **Secondary Metabolites**         | **Concentration** |
| Phenols                           | 110.0 ± 0.44 mg/g gallic equivalent |
| Flavonoids                        | 16.24 ± 0.48 mg/g quercetin equivalent |
| Alkaloids                         | 3.637 ± 0.028 mg/ml |
| Saponins                          | 6.819 ± 0.024 mg/ml |
| Tannins                           | 1.855 ± 0.006 mg/ml |
| Glycosides                        | 0.3178 ± 0.003 mg/ml |
| Terpenoids                        | Not detected |
| Anthraquinones                    | Not detected |
| Volatile oil                      | Not detected |

Note: Data are means ± SEM of three replicates.

3.2. Acute toxicity of A. nilotica pod

Oral acute administration of 3,000 mg/kg bw of A. nilotica seedpod aqueous extract resulted in no mortality, suggesting that the 

3.3. Anti-ulcerogenic activities of A. nilotica seedpod aqueous extract

Tables 2a and 2b show the gastric secretory indices of indomethacin and ethanol-ulcerated rats orally administered A. nilotica seedpod aqueous extract for seven days. Gastric juice pH in ulcerated rats induced with indomethacin was (p < 0.05) reduced, whereas the gastric volume, free and total acidity were increased when compared to the control. A. nilotica seedpod aqueous extract treatment for seven days to indomethacin-ulcerated rats normalized the pH and reduced the volume and acidity of gastric secretion significantly (p < 0.05) from the control. Pepsin level in the gastric juice in both indomethacin and ethanol-induced rats were also increased in comparison to the control group. Treatment with 50 mg/kg bw of the extract significantly (p < 0.05) reduced this in both ulcer healing and gastroprotective studies.

| Note: Data are means of six replicates ± SEM. Values with different superscript down the same column are significantly different (p < 0.05); ANP: Acacia nilotica seedpod. |
### Table 2b: Gastric acid secretory indices in ethanol - Induced ulcerated rats treated with *A. nilotica* seedpod aqueous extract

| Treatment               | Gastric pH (±SEM) | Gastric volume (mL) | Free acidity (Meq/L/100g) | Total acidity (Meq/L/100g) | Gastric pepsin (mg/mL) |
|-------------------------|-------------------|---------------------|---------------------------|----------------------------|------------------------|
| Control                 | 3.93 ± 0.17        | 1.53 ± 0.14         | 9.6 ± 0.75                | 16.33 ± 1.17               | 2.67 ± 0.47           |
| Ulcerated               | 4.30 ± 0.08        | 2.53 ± 0.15         | 2.67 ± 0.80               | 7.83 ± 1.05                | 4.80 ± 0.11           |
| Ranitidine (10 mg/kg)   | 3.67 ± 0.34        | 0.82 ± 0.16         | 4.17 ± 1.28               | 11.33 ± 1.43               | 4.73 ± 0.13           |
| ANP (12.5 mg/kg)        | 3.38 ± 0.21        | 1.40 ± 0.15         | 3.0 ± 0.45                | 12.0 ± 1.57                | 4.69 ± 0.15           |
| ANP (25 mg/kg)          | 3.95 ± 0.09        | 1.87 ± 0.13         | 4.83 ± 0.80               | 19.83 ± 1.96               | 3.22 ± 0.07           |
| ANP (50 mg/kg)          | 3.73 ± 0.17        | 2.00 ± 0.19         | 4.67 ± 0.62               | 17.17 ± 1.47               | 1.21 ± 0.09           |

**Note:** Data are means of six replicates ± SEM. Values with different superscript down the same column are significantly different ($p < 0.05$); ANP: *Acacia nilotica* seedpod.

*A. nilotica* extract significantly ($p < 0.05$) decreased ulcer index in a dose-dependent manner as shown in Table 3a. Highest reduction in ulcer index was observed in rats treated with 50 mg/kg bw of the extract resulting in 70.25% healing of indomethacin-induced ulceration (Table 3a); the reduction was the same as that of ANP.

### Table 3a: Effect of *A. nilotica* seedpod aqueous extract on gastric wall indices in indomethacin - Induced ulcerated rats

| Treatment               | Ulcer Index (Meq/L/100g) | Ulcer healing (%) | Adherent Mucin (mg/mL) |
|-------------------------|---------------------------|-------------------|------------------------|
| Control                 | _                         | _                 | 115.77 ± 1.49          |
| Ulcerated               | 26.33 ± 0.60              | _                 | 65.95 ± 0.60           |
| Ranitidine (10 mg/kg)   | 7.5 ± 0.50                | 71.32             | 88.53 ± 3.70           |
| ANP (12.5 mg/kg)        | 22.0 ± 0.78               | 16.45             | 72.76 ± 2.79           |
| ANP (25 mg/kg)          | 16.33 ± 0.56              | 37.98             | 72.04 ± 2.36           |
| ANP (50 mg/kg)          | 7.33 ± 0.70               | 70.25             | 99.25 ± 3.43           |

**Note:** Data are mean of 6 replicates ± SEM; values with different superscript are significantly different ($p < 0.05$); ANP: *Acacia nilotica* seedpod.

### Table 3b: Effect of *A. nilotica* seedpod aqueous extract on gastric wall indices in ethanol - Induced ulcerated rats

| Treatment               | Ulcer index (Meq/L/100g) | Ulcer healing (%) | Adherent Mucin (mg/mL) |
|-------------------------|---------------------------|-------------------|------------------------|
| Control                 | _                         | _                 | 78.17 ± 0.76           |
| Ulcerated               | 21.17 ± 0.54              | _                 | 32.15 ± 0.08           |
| Ranitidine (10 mg/kg)   | 1.83 ± 0.48               | 91.29             | 23.26 ± 0.10           |
| ANP(12.5 mg/kg)         | 14.83 ± 1.01              | 29.38             | 27.31 ± 0.12           |
| ANP(25 mg/kg)           | 20.0 ± 0.37               | 4.76              | 24.52 ± 1.04           |
| ANP(50 mg/kg)           | 12.00 ± 0.73              | 42.86             | 26.31 ± 0.99           |

**Note:** Data are mean of 6 replicates ± SEM; values with different superscript are significantly different ($p < 0.05$); ANP: *Acacia nilotica* seedpod.
those treated with the referenced drug (ranitidine). Also, a gastroprotective study showed that pretreatment with 50 mg/kg bw of \textit{A. nilotica} aqueous extract for seven days, followed by ulcer induction with ethanol caused a significant \((p < 0.05)\) reduction in ulcer index resulting in 42.86% inhibition of ulceration, though ranitidine showed a better 91.29% ulcer inhibition (Table 3b). Induction of ulceration in rats significantly reduced the concentration of adherent mucin. Rats treated with the extract showed a significantly \((p < 0.05)\) higher amount of adherent mucin than indomethacin-ulcerated rats. However, treatment with the extract did not ameliorate the significant reduction in the level of adherent mucin in ethanol-induced ulcerated rats.

Tables 4a and 4b show the effect of \textit{A. nilotica} Seedpod Aqueous Extract on some biochemical parameters on gastric mucosa of ulcerated rats. The activity of myeloperoxidase was significantly \((p < 0.05)\) increased following induction of ulcer with either indomethacin or ethanol. Administration of ranitidine or \textit{A. nilotica} extract significantly \((p < 0.05)\) reduced the activity of the enzyme. Malondialdehyde (MDA) concentration was significantly \((p < 0.05)\) elevated in the ulcer-induced rats when compared to normal control, this was however

### Table 4a: Effect of aqueous extract of \textit{A. nilotica} seedpod on some biochemical parameters in gastric mucosa of indomethacin-induced ulcerated rats

| Treatment            | MPX (nmol/min/ml) | MDA (µmol/mg protein) | GST (µmol/min/ml) | GPx (µmol/min/ml) | GSH (µmol/ml/min) | SOD (nmol/min/ml) | Catalase (nmol/min/ml) |
|----------------------|-------------------|------------------------|-------------------|-------------------|-------------------|-------------------|------------------------|
| Control              | 0.192 ± 0.011      | 0.968 ± 0.009          | 0.165 ± 0.002     | 1.750 ± 0.010     | 26.966 ± 0.328    | 17.708 ± 0.228     | 9.798 ± 0.126          |
| Ulcerated            | 0.271 ± 0.007      | 1.320 ± 0.041          | 0.036 ± 0.007     | 0.159 ± 0.010     | 21.320 ± 0.166    | 12.459 ± 0.144     | 6.442 ± 0.116          |
| Ranitidine (10 mg/kg)| 0.169 ± 0.005      | 1.071 ± 0.054          | 0.082 ± 0.005     | 1.619 ± 0.048     | 26.260 ± 0.353    | 16.559 ± 0.080     | 9.560 ± 0.265          |
| ANP (12.5 mg/kg)     | 0.191 ± 0.006      | 0.602 ± 0.052          | 0.106 ± 0.001     | 0.583 ± 0.031     | 25.760 ± 0.128    | 13.585 ± 0.071     | 9.631 ± 0.318          |
| ANP (25 mg/kg)       | 0.175 ± 0.001      | 0.556 ± 0.016          | 0.114 ± 0.003     | 0.320 ± 0.014     | 22.544 ± 0.597    | 16.509 ± 0.093     | 9.247 ± 0.187          |
| ANP (50 mg/kg)       | 0.157 ± 0.001      | 0.479 ± 0.027          | 0.122 ± 0.006     | 0.715 ± 0.094     | 23.147 ± 0.884    | 15.709 ± 0.157     | 9.401 ± 0.270          |

**Note:** Data are mean of 6 determinations ± SEM. Values with different superscript down the same column are significantly different \((p < 0.05)\). MPX: Myeloperoxidase; MDA: Malondialdehyde; GST: Glutathione transferase; GPx: Glutathione peroxidase; GSH: Glutathione; SOD: Superoxide dismutase; ANP: \textit{Acacia nilotica} seedpod.

### Table 4b: Effect of aqueous extract of \textit{A. nilotica} seedpod on some biochemical parameters in gastric mucosa of ethanol-induced ulcerated rats

| Treatment            | MPX (nmol/min/ml) | MDA (µmol/mg protein) | GST (µmol/min/ml) | GPx (µmol/min/ml) | GSH (µmol/ml/min) | SOD (nmol/min/ml) | Catalase (nmol/min/ml) |
|----------------------|-------------------|------------------------|-------------------|-------------------|-------------------|-------------------|------------------------|
| Control              | 0.223 ± 0.006     | 0.886 ± 0.047          | 0.077 ± 0.003     | 2.232 ± 0.020     | 20.400 ± 0.189    | 24.140 ± 0.522     | 12.417 ± 0.095          |
| Ulcerated            | 0.423 ± 0.011     | 0.847 ± 0.025          | 0.007 ± 0.006     | 1.525 ± 0.091     | 17.863 ± 0.543    | 17.203 ± 0.529     | 6.379 ± 0.205          |
| Ranitidine (10 mg/kg)| 0.112 ± 0.021     | 0.753 ± 0.025          | 0.038 ± 0.002     | 2.733 ± 0.053     | 22.963 ± 0.755    | 30.383 ± 0.695     | 8.801 ± 0.031          |
| ANP (12.5 mg/kg)     | 0.196 ± 0.003     | 0.947 ± 0.044          | 0.029 ± 0.008     | 2.588 ± 0.042     | 21.827 ± 0.980    | 28.283 ± 1.165     | 10.223 ± 0.001          |
| ANP (25 mg/kg)       | 0.165 ± 0.004     | 0.858 ± 0.016          | 0.145 ± 0.002     | 2.596 ± 0.079     | 20.310 ± 1.107    | 24.16 ± 0.376      | 5.621 ± 0.204           |
| ANP (50 mg/kg)       | 0.157 ± 0.001     | 0.850 ± 0.041          | 0.168 ± 0.005     | 3.083 ± 0.055     | 23.147 ± 1.336    | 20.897 ± 0.643     | 13.933 ± 0.134          |

**Note:** Data are mean of 6 determinations ± SEM. Values with different superscript down the same column are significantly different \((p < 0.05)\). MPX: Myeloperoxidase; MDA: Malondialdehyde; GST: Glutathione transferase; GPx: Glutathione peroxidase; GSH: Glutathione; SOD: Superoxide dismutase; ANP: \textit{Acacia nilotica} seedpod.
lowered in groups treated with the aqueous extract of *A. nilotica* and ranitidine. Glutathione transferase, glutathione peroxidase, superoxide dismutase, and catalase activities were increased significantly (p < 0.05) in *A. nilotica* treated groups when compared to ulcerated rats. Glutathione levels in animals treated with indomethacin and ethanol were significantly (p < 0.05) reduced than normal control, this was brought towards normalization in *A. nilotica* treated ulcerated rats.

### 4. Discussion

There has been a growing recognition of the therapeutic usefulness of medicinal plants in recent years (Yadav and Agarwala, 2011). This renewed interest has made researchers more concerned with the validation of traditional usage of plants including identification, isolation, and characterization of active components (Fennell et al., 2004). Global pharmaceutical industries are now looking for better drug options that will reduce the innovation deficit (Romulo and Joao Paulo, 2011). The effectiveness and mechanisms of action of crude herbal extracts vary according to their active constituents (Bhavani et al., 2012). These compounds are synthesized by the secondary metabolism of plants, they are diverse compounds with a lot of known and unknown functions (Yadav and Agarwala, 2011).

Alkaloids present in *A. nilotica* have been shown in previous studies to possess exhibit antiulcer effect (Tan and Nyasse, 2000). Saponins are found in different amount in plants and possess antiulcerogenic activity (Maity and Chattopadhyay, 2008). They act mainly by inhibiting the secretion of gastric acid and lowering gastric juice pH (Lee et al., 2005). Tannins help in the precipitation of mucosa proteins thus inhibiting the penetration of irritants that could cause injury (Thirunavukkarasu et al., 2009).

Flavonoids are phenolic compounds, they also gastroprotective action in mammals (Awaad et al., 2013). The high content of total phenolic compounds found in *A. nilotica* seedpod may account for the anti-ulcerogenic properties observed in the present study.

It is important to establish a safety profile of *A. nilotica* which could be used as a guide for its use in herbal medicine. This helps to prevent the occurrence of any toxicity risk that may arise from using *A. nilotica*. Such risks are assessed by using the compound at varying doses and different duration (Schultz et al., 2001).

According to Partrick-iwuanyanwu et al. (2012), a material with toxicity greater than 1000mg/ kg body weight may be considered of low toxicity and can be used safely. In the present acute toxicity study (LD₅₀) single administration of 3,000 mg/ kg body weight of *A. nilotica* shows no toxic sign. This result suggests that *A. nilotica* is not toxic after acute exposure of not more than 3,000 mg/ kg bw in rats.

Vasconcelos et al. (2008) have suggested that both ulcer healing and gastroprotective activity of substances needed to be affirmed. The use of Non-Steroidal Anti-inflammatory Drugs (NSAIDs) like aspirin and indomethacin over long periods during anti-inflammatory therapy induces gastric ulcers (Wallace, 2001). Alcohol is also a risk factor for gastric ulcer development, it readily penetrates the gastric mucosa and causes infiltration of inflammatory cell, lipid peroxidation that ultimately leads to gastric mucosal lesions (Dinortey et al., 2013). The ethanol induction model was used for the gastroprotective study of *A. nilotica* seedpod while the indomethacin-induced ulceration model was used to study the ulcer healing properties of *A. nilotica* seedpod.

Gastric acid secretion has been largely implicated in the development and healing of ulcers and control of its secretion is important for ulcer healing and prevention. Ranitidine is a histamine receptor antagonist, it blocks the H₂ receptor resulting in a decrease in the secretory actions of gastrin and acetylcholine on parietal cells (Norlin et al., 2005). A previous study on the young seedless pod of *A. nilotica* shows that it has significant antisecretory activity in pylorus-ligation-induced ulcers (Bansal and Goel, 2012). The changes observed in gastric secretions of rats treated with aqueous extract of *A. nilotica* seedpod suggest that it has an anti-secretory effect on the parietal cells in both indomethacin and ethanol ulcerated rats. Gastric mucus also protects the gastric mucosa mainly because of its glycoprotein (mucin) content (Oyegoke and Oladiji, 2014). The increased mucin secretion of treated animals suggested that the antiulcerogenic activity elicited by *A. nilotica* seedpod is probably due to the enhancement of gastric mucosa defense.

Ulcer index is a visible indicator of gastric erosion or injury used to evaluate the degree of ulceration (Shuai et al., 2011). *A. nilotica* seedpod enhanced the mucosa structural integrity thus reducing the formation of lesions on the glandular part of the mucosa. The results also suggested that *A. nilotica* seedpod will be a better agent for ulcer healing than a gastroprotective agent as evidenced by the higher percentage of ulcer healing.
Hydroethanolic extract of young seedless pods of A. nilotica has been reported to exhibit antiulcer activity in different ulcer rat models (Bansal and Goel, 2012).

Myeloperoxidase is a heme-containing enzyme found in lysosomes (Bhattacharyya et al., 2014). Its activity is an indication of cells’ inflammatory responses in various clinical conditions (Devi et al., 2007). Increased activity of myeloperoxidase was observed in the ulcerated rats indicating an increased level of activated neutrophil due to the injurious effect of the ulcerogens on the mucosa cells. The rats treated with the aqueous extract of A. nilotica showed a reduced myeloperoxidase activity indicating that it is a potent anti-inflammatory agent.

Reactive oxygen species (ROS) and reactive nitrogen species (RNS) also have a role in gastric ulcer formation (Zakaria et al., 2014). This harmful effect is prevented by antioxidants, these comprise both enzymatic and nonenzymatic antioxidants (Bhattacharyya et al., 2014). There was a significant increase in the concentration of products of lipid peroxidation in form of malondialdehyde in stomach tissues administered with indomethacin and ethanol, this was however decreased in groups treated with the aqueous extract of A. nilotica.

Natural food-based antioxidants have been shown to delay and inhibit the onset of gastric ulcers and carcinoma (Panneerselvam and Arumugam, 2011). Previous reports have shown that the administration of NSAID and ethanol decrease the activity of superoxide dismutase, glutathione peroxidase, and the level of reduced glutathione (Bafna and Balaraman, 2004). Administration of A. nilotica seedpod aqueous extract however increased the levels of reduced glutathione. Reduced glutathione is needed by glutathione-dependent antioxidants; glutathione peroxidase and glutathione transferase. Its non-availability makes these enzymes action to be below optimum (Devi et al., 2007). The extract-treated groups had higher concentration of glutathione, possibly because it served as an antioxidant thus reducing the consumption of gastric glutathione. These results show that A. nilotica seedpod aqueous extract has an antioxidant effect.

5. Conclusion

This study has shown that A. nilotica seedpod exhibited both gastroprotective and ulcer healing effects by the up-regulation of the mucosal defense system and stimulation of the antioxidant defense system hence it could be recommended as a good candidate for the development of cheaper therapeutic options for PUDs.

Declaration of Competing Interest

The authors declare no conflict of interest.

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