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Research article

Effect of an Aqueous Extract of *Entandrophragma utile* Bark on Gastric Acid Secretion in Rat and Isolated Ileum Contractility in Guinea pig

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ABSTRACT: Adjunct therapy is needed for patients with compromised gastrointestinal mucosa due to necessary aspirin usage against cardiovascular disorders. We tested the Nigerian bark extract of *Entandrophragma utile* on gastric acid secretion (GA) and peptic activity (PA). Rats were ligated at the pylorus for collection of gastric juice which was measured for PA spectrophotometrically using bovine serum albumin as substrate and for titratable GA using phenol red indicator. The extract was compared with ranitidine (histamine H₂-receptor antagonist). The extracted was tested on isolated guinea-pig ileum preparations for histaminergic responses and was compared with mepyramine (histamine H₁-antagonist). *E. utile* reduced GA to 1.33 ± 0.6 uEq g⁻¹ from 2.82 ± 0.7 uEq g⁻¹ in controls using 6h ligations. For 4h ligations, control PA (mg/dL BSA digested) was 38.75 ± 4.05 which was lowered to 14.8 ± 4.67 (p<0.01) by the extract and to 3.4 ± 0.72 (p<0.001) by ranitidine. Chronic administration of *E. utile* decreased GA in 4h collections. *E. utile*, 10 -160 x 10⁻³ g, antagonized 10 µg histamine-induced contractions by 28-62% dose-dependently. Mepyramine gave a parallel shift of the histamine dose-response graph to the right typical of a competitive antagonism. *E. utile* extract gave a non-parallel shift to the right with a lowering of maximal response typical of a non-competitive antagonism. The two properties of the *E. utile* extract on acid and pepsin may be valuable for patients on aspirin with compromised mucosa therefore the extract could be developed as adjunct therapy to minimize aggravation of mucosal damage by acid-peptic autodigestion.

Keyword: *Entandrophragma utile*, peptic ulcer, stomach acid, peptic activity, antisecretory, Shay rat

INTRODUCTION

It has long been known that gastric acidity and enzymatic activity of pepsin are major culprits generating ulceration in the compromised gastrointestinal mucosa (Gunzburg, 1852, Beaumont, 1883). Hypersecretory states may lead to autodigestion of the gastric mucosa and painful ulceration. In fact, Quincke (1882) introduced the term “peptic ulcer” to describe ulcers of the gastrointestinal tract. Duodenal

ulcers are especially associated with hypersecretory states (Richard, 1985). Treatment of peptic ulcers aims at buffering defense and integrity of the mucosa and limiting the influence of acid and pepsin. Both the hydrochloric acid and pepsinogen secreted by the gastric glands (McMinn and Hobdell, 1974) are important for normal digestive function. However, for ulcer patients, it may be desirable to inhibit such secretion. In recent times the use of aspirin as an important cardio-protective, especially in elderly persons, has surged because it has been observed to prevent stroke, some forms of angina, vascular thrombosis as well as other disease conditions. Aspirin consumption was 40 billion tablets per annum according to a 1989 report by Barrier and Hirshchowitz. Non-steroidal anti-inflammatory drugs (NSAIDs) such as aspirin have been known to cause peptic ulcers for a long time (Lang, 1957; Levrat and Lambert, 1960). Aspirin, both by being a COX-1 inhibitor and a generator of toxic oxygen metabolites, compromises gastrointestinal mucosal integrity. A new breed of peptic ulcer patients have been generated by

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the increased use of aspirin therefore there is need for development of adjunct therapy to prevent the ulcerogenic side effect of this important drug (Anand *et al.*, 1999). The aqueous bark extract of *Entandrophragma utile* is used in Nigerian traditional medicine to treat gastrointestinal ulcers (John and Onabanjo, 1990a and 1990b, John 1994). This report shows its effect in inhibiting gastric acidity and peptic activity in Shay rats.

MATERIALS AND METHODS

Collection and extraction of plant material

Fresh pieces of the bark were obtained from a tree in its natural habitat at Ojokoro, on the outskirts of Lagos, Nigeria with the help of an experienced traditional herbalist, Chief E. O. Esho. The botanical profile was confirmed at the Forestry Research Institute of Nigeria, Ibadan with the support of a taxonomist, Dr. M. O. Soladoye. The fresh bark was used within a week of collection. If not used on the day of collection, it was kept in refrigeration at 4°C. Before use, the bark was chopped into pieces of about 1 cm³ and rinsed properly in water. According to Chief Esho, about 200 g of chopped bark is simmered for 2 h in about 1L water at about 95°C. We adapted this to glass Soxhlet flask extraction which allows a constant volume of solvent to be used throughout the extraction and this improves calculation of both concentration and yield of extract from the original starting materials measured with precision each time.

The extract was then decanted and filtered while hot with Whatman filter paper No. 1 and then cooled to room temperature (30 + 2°C) in a stoppered flask. The volume of the extract was measured and the pH determined using a pH meter (Seibolo Wien, Australia). The used bark was then dried and re-weighed for calculating the extract derived from it. The concentration of the extracts so obtained in the procedures were calculated as a difference in the weight of starting and used bark material per millilitre, (g ml⁻¹). The tannin contents of the extract gave it a reddish brown coloration and this posed a problem for phenol red assay of acidity therefore we decolorized the extract although we did not know if the tannic content contributed to any pharmacological effect of the extract. Activated charcoal (5 g) was mixed with 1 L of the extract and this was then stirred for 15 min over a hot plate (50 °C). It was left at room temperature for 1 h to allow binding of tannins. It was then filtered hot with Whatman filter paper No. 1. The filtrate was re-filtered, if necessary, using Whatman filter paper No. 5 until the charcoal had completely disappeared from the

filtrate. The resultant filtrate was stored in stoppered flasks in a refrigerator at 4°C for not more than two weeks after which the extracts were discarded and fresh ones prepared. The doses of extract given were expressed in gram per kilogram body weight (g kg⁻¹).

Animals

Male and female Sprague-Dawley rats weighing 200 ± 20 g were obtained from the Laboratory Animal Centre (LAC), College of Medicine, University of Lagos (CMUL) and utilized according to ethical approval by the LAC. Food was withdrawn overnight before experiments. During food withdrawal, animals had access to water *ad libitum* and were kept in wide-mesh bottom cages to prevent coprophagy. They were sex and weight matched between groups per experiment.

Shay rat preparations

Anesthetized rats were ligated at the pylorus as described by Shay *et al.* (1954) in order to collect secreted gastric acid and pepsin. Each rat was anaesthetized lightly in an ether chamber. A midline incision of about 2cm extending from the xiphoid was made on the ventral abdominal wall to expose the abdominal cavity. A ligature was placed at the junction between the pylorus and duodenum with care being taken to avoid damage to the gastro-duodenal artery or traction on the stomach. A No. 8 polyethylene tube was carefully passed through the oesophagus into the stomach with the tip impinging on the stomach wall. Gastric lavage was carried out through the tube to remove food residues and gastric secretions. Four milliliter of physiological saline was injected and withdrawn immediately by gentle suction with a 5ml syringes while gradually withdrawing the tube. The tube was passed the second time into the stomach to recover any fluid that might still be there, and was subsequently removed (a total of 4ml or more was recovered). The opened abdominal wall was immediately closed by suturing and the area cleaned with physiological saline to remove blood stain. Transparent lacquer (nail varnish) was used to coat the dried sutured surface area to prevent leakage and ingestion of body fluids during grooming by the rat which invariably may affect the analysis of subsequent gastric effluents. After the operation, 5ml of 0.9% NaCl was injected subcutaneously (s.c.) to make up for the loss of body fluid during the process of the operation. The rat was left in a clean cage without food, water or ingestible materials to recover from the anaesthesia. After 4h accumulation of gastric secretion, the rat was sacrificed by stunning once. The abdomen was again opened and a ligature placed on the oesophago-cardiac junction to prevent leakage of the

acid effluent during excision of the stomach from the oesophagus and the duodenum. The stomach was then removed, washed in physiological saline and dried with filter paper. An opening was thereafter made along the greater curvature through which the gastric juice was drained into a 10ml measuring cylinder with a funnel.

Analysis of gastric acid content

The gastric contents of each stomach were analyzed individually. The contents were placed in centrifuge tubes and centrifuged at 5000g for 20min in a Minor Centrifuge (ESE, England) at room temperature (30°C). After centrifuging, the supernatant fluid volumes (V_E) and solid residues were recorded. If the residue measured over 0.6 ml in the tube, the preparation was discarded because of contamination of the gastric effluent by coprophagy. The titratable acidity in the supernatant fluid was then determined. Aliquots of 0.5 or 1ml of the supernatant fluid (depending on the quantity obtained) were titrated against 0.1N NaOH using phenolphthalein indicator. The titration was duplicated where the volume of gastric effluent was adequate. The titratable acidity was calculated as in simple volumetric analysis, and expressed finally as micro-equivalent of titratable acid (H^+) per kilogram fasting body weight of rat per 4h ($\mu E \text{ kg}^{-1} 4h^{-1}$).

Peptic activity measurement

Using the method of Prino *et al.* (1971) the enzymic activity of pepsin in undiluted gastric juice was measured using bovine serum albumin (BSA) as substrate. The principle is based on the proteolysis of the albumin substrate by gastric juice resulting in the production of peptides which contain tyrosine and tryptophan whose relative density measured spectrophotometrically are indicative of the concentration of pepsin in the gastric juice. Conversely, a reduction in the peptides represents an inhibition of proteolysis (antipepsin activity).

The gastric effluent collected was first centrifuged at 5000g for 10 min. A sample of 0.1 ml of the supernatant fluid was added to 1 ml of 0.5% BSA in 0.01N HCl (pH 2). The gastric juice sample and BSA were mixed well and incubated in a water bath (37°C) for 20 minutes. A control cuvette containing gastric juice was prepared and albumin was replaced with 1ml 0.01N HCL. The hydrolysis was brought to a stop after 20 min incubation by adding 2ml of 10% trichloroacetic acid (TCA). All tubes were heated in boiling water for 5 min, cooled and filtered through Whatman No.42 filter paper. To a sample of 2ml of the filtrate was added 0.8ml of 2.5N NaOH and 0.2ml Folin-Ciocalteu reagent. The volume was made up to 20ml with distilled water. The absorbency of the

protein was read in a Pye Unicam SP6-450 UV/VIS spectrophotometer (Phillips, England) at 700 nm.

The second method used for the measurement of peptic activity from the gastric contents of the stomach of pylorus-ligated rats was that reported by Ohnishi and Barr (1978). The following reagents were used: Reagent A or biuret reagent (Sigma U.S.A) contained 0.75 mmol l^{-1} C_6SO_4 , 94 mmol l^{-1} NaOH, tartarate, iodide and carbonate; Reagent B or Folin-Ciocalteu phenol reagent (Sigma, U.S.A) (this prepared reagent was standardized by the supplier and contained 2N of Folin reagent); Reagent C or bovine serum albumin (Sigma U.S.A) was the standard protein prepared as 1.5 mg ml^{-1} BSA in 0.01N HCl.

The preparation of the gastric acid filtrate up to hydrolysis was as in the Prino *et al.* method above and was used as the starting material in the present determination. To each cuvette was added 0.2ml gastric acid filtrate and 2.2 ml of reagent A. The tube contents were mixed thoroughly and left to stand at room temperature for 10 min. After this, 0.1ml of reagent B was added and the tube contents were mixed immediately and left to stand for 30 min or longer at room temperature for colour development. The blank cuvette contained 0.2ml saline instead of the gastric acid filtrate and the other reagents were added as for test cuvettes.

The absorbency of the contents in the cuvettes were read in a Pye Unicam SP6-450 UV/VIS spectrophotometer (Phillips, England) at 700 nm. The protein concentration was calculated from a standard curve. The test and blank were determined in duplicate.

Standard curve

The standard curve for protein determination was obtained by reconstituting in physiological saline reagent C which served as the stock solution. The latter was diluted in concentrations of 0, 25, 50, 75 and 100 mg l^{-1} of total protein. The prepared concentrations of reagent C were mixed similarly as above with reagents A and B in 3 ml cuvettes. The optical density was read at 725 nm and plotted against protein concentration. The protein concentrations of gastric juice samples were determined from the curve and used for the basic calculation of peptic activity.

Test of the effect of *E. utile* extract or ranitidine on 4 h gastric acid output (GAO) and peptic activity (PA)

Peptic activity (PA) and gastric acid output (GAO) were determined as above. Determination of PA was carried out before GAO measurement. PA was measured by both the method of Prino *et al* (1971) and the method of Ohnishi and Barr (1978).

The three tests just described were run simultaneously using weight-matched rats on each occasion as follows: two rats for control given 1 ml saline orally, two for treatment with 20 mg ranitidine in 1 ml orally, and two for treatment with 1ml of 100 mg ml⁻¹ extract of *E. utile* given orally. Saline or test agent was given immediately after ligation and animals were sacrificed 4h post-ligation. The results of six determinations were pooled at the end of the series.

Evaluation of gastric acid output for varying ligation periods in rats treated with the *E. utile* extract

Six male weight-matched rats were used in each determination. Three were used as controls and were given 1 ml saline orally after ligation. The other three were given orally 1ml of 100 mg ml⁻¹ extract of *E. utile* immediately after ligation. They were kept for 3, 5 and 6 h post-ligation respectively before being sacrificed. The GAO measurement of gastric effluents was as described above. The experiment was repeated for pooled sextuplicate data.

Investigation of the effect of six weeks administration of *E. utile* extract on GAO and PA

Twenty rats were fed and kept normally for six weeks during which they were given a suspension of 100 mg ml⁻¹ aqueous extract of *E. utile* to drink *ad libitum* in their feeding bottles. Fresh extracts were supplied in their bottles daily in place of water. The rats were observed to drink freely and did not show any sign of displeasure to the drink. After six weeks, the rats were tested for GAO as above. The PA determination was carried out by the Prino *et al.* (1971) method.

Investigation of the effect of *E. utile* extract on histaminergic contractions of the guinea-pig ileum.

Guinea-pigs of either sex weighing 250-300g were used. Each animal was sacrificed by a single stun and bled at the jugular vein. A piece of about 10cm long of the terminal ileum was excised. The lumen was washed out with Tyrode solution with a pipette. A 2-3cm segment was cut off and suspended in a 5ml bath of Tyrode solution. The organ bath was maintained at 35°C and aerated with a constant flow of oxygen. The lower end of the isolated tissue was attached by means of a looped thread to a hook on the glass oxygen tube. The upper end was hooked to a force transducer mounted and connected to a 2-Channel "Gemini" electronic recorder (Ugo Basile, Italy) to record contractions. The tension on the tissue was 10g. Test substances were left in contact with the tissue for thirty seconds before being washed out three times by upward displacement of bathing fluid.

Measurement of the effect of the aqueous extract of *E. utile* on guinea-pigs ileum

Doses of 2-10 x 10⁻³ g of aqueous *E. utile* extract were added to the organ bath alternating with 1-10-ug histamine acid phosphate. Recordings were made using a dose cycle of 3min with 20s contact time of the extract with the ileum.

Determination of histamine responses with increasing doses of *E. utile* extract

The bathing Tyrode solution had 2 x 10⁻⁶g L⁻¹ atropine added to block cholinergic receptors. A bracketing assay was done using a dose of 10 ug histamine. The extract, added to the organ bath 30s before histamine, was used as the antagonist and the response recorded. The response to histamine alone was again recorded. The sequence was repeated using graded doses of the extract each time 10 -160 x 10⁻³ g with the constant dose of histamine (10 ug).

Investigation of anti-histamine effect of the extract of *E. utile*

The ileum was bathed with atropinised Tyrode solution as above. Contractions of the tissue to 0.002 - 0.256 mg histamine were recorded to obtain a dose-response relationship. Then 5 x 10⁻⁶ M mepyramine, an H₁ receptor-antagonist was also added before repetition of the histamine graded responses. Similarly, the *E. utile* extract (10 x 10⁻³ g) was added in place of mepyramine and the histamine responses were repeated. Ranitidine (40 ug) (Glaxo, Nigeria Ltd), an H₂ receptor-antagonist was also added to the organ bath and the histamine responses were recorded.

RESULTS

The aqueous bark extract of *E. utile*

The aqueous bark extractions obtained were reddish-brown with a faint pleasant scent and a pH of 3 to 4.4. After treatment of the extract with activated charcoal, the recovered extract was pale yellow and had a pH of 4 ± 0.2.

Effect of *E. utile* extract on gastric acid in the Shay rats

The total amount of titratable gastric acid output (GAO) produced by each rat for a particular period was calculated using total volume of the clear gastric effluents (V_E) and the titre value. In Figure 1A, which depicts 4h ligation measurements, ranitidine significantly decreased titratable acid (p

< 0.005). The stomach effluents of *E. utile* treated rats did not show lower acidity than controls. Figure 2 shows the relationship between GAO and ligation period. Titratable acidity was lower than controls in *E. utile* treated rats in the 6h ligation group ($p < 0.05$) and not in shorter ligation periods. Control values obtained with mean GAO were 0.39 ± 0.1 , 0.38 ± 0.2 , 0.61 ± 0.21 and 2.82 ± 0.7 $\mu\text{Eq g}^{-1}$ for 3, 4, 5 and 6h ligation respectively. With the administration of *E. utile* extract, the

mean GAO values were 0.55 ± 0.2 , 0.53 ± 0.17 , 0.59 ± 0.13 and 1.33 ± 0.6 respectively for 3, 4, 5 and 6h ligation.

The levels of significance for the difference between controls and extract treated rats were $p < 0.05$ for 3h period, $p > 0.05$ for 4 and 5 h ligations and $p < 0.01$ for 6h ligation. Chronic administration of *E. utile* extract resulted in block of gastric acid production (Figure 3) using the 4h ligation period.

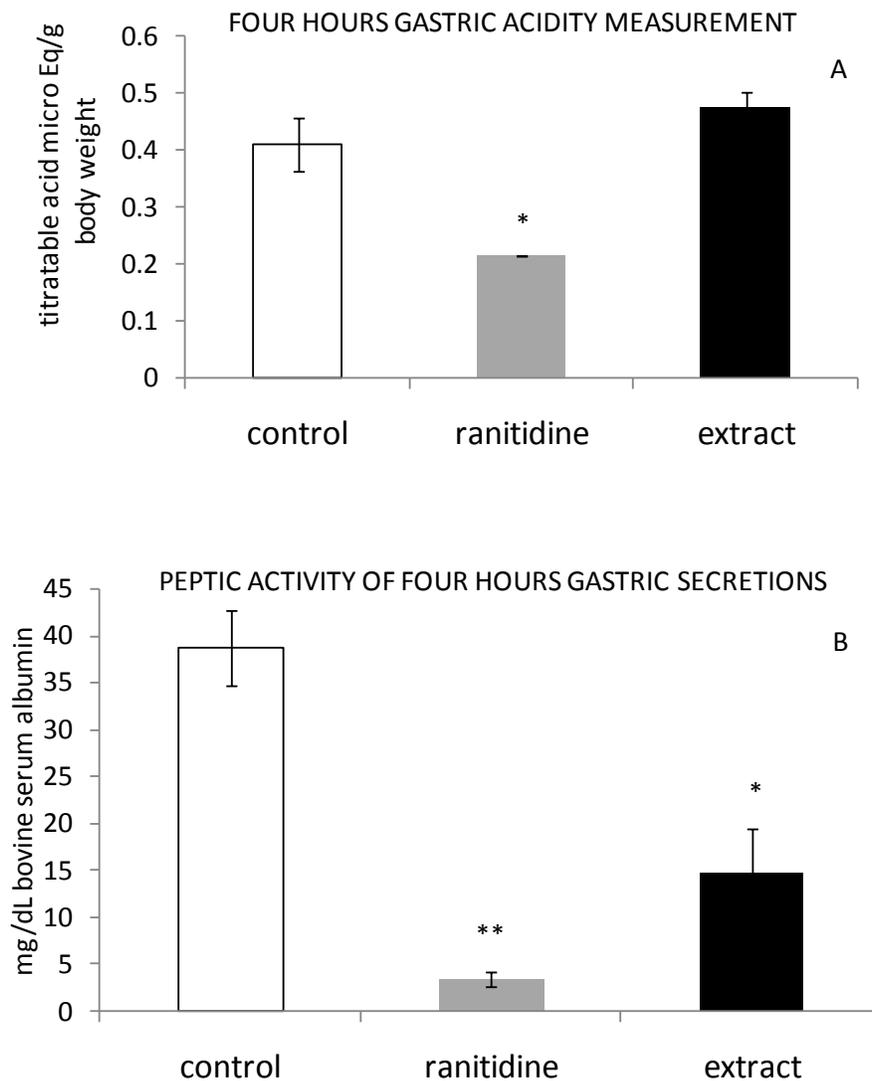


Fig. 1

Results of analysis of gastric effluents collected over 4h after ligation of rat pylori. Drugs were given orally immediately after ligation. Gastric effluents were collected from sacrificed animals, centrifuged, and analysed for titratable acidity using phenol red and for peptic activity by spectrophotometrical assay of peptic digestion of bovine serum albumin. Ranitidine, a clinically effective anti-ulcer drug, significantly decreased titratable gastric acidity ($p < 0.005$) and peptic activity of gastric effluents ($p < 0.001$). The *E. utile* extract significantly decreased peptic activity during 4h ($p < 0.01$).

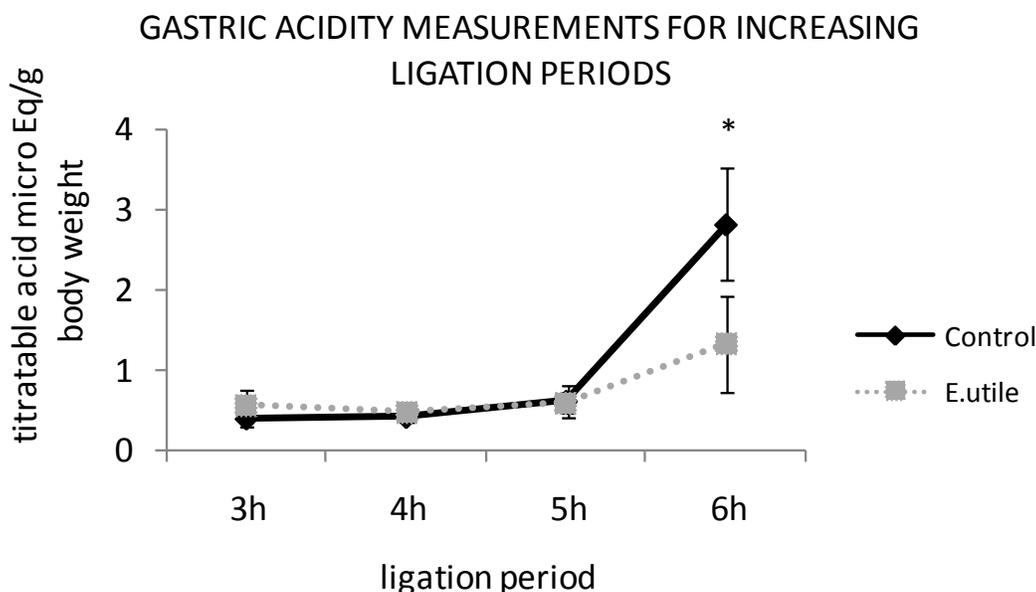


Fig. 2

Acute administration of *E. utile* bark extract negatively affects gastric acid secretory mechanisms. Control rats were given 1 ml saline and test rats were given 1ml of 100 mg ml⁻¹ *E. utile* extract by oral cannulation immediately after ligation. Inhibition of acid secretion was observable by the Shay rat acid titration method when the ratio secreted gastric acid/aqueous extract content of stomachs is high (6h ligation). The significance of the difference between the controls and the extract treated stomachs for the 6h ligation is p<0.01.

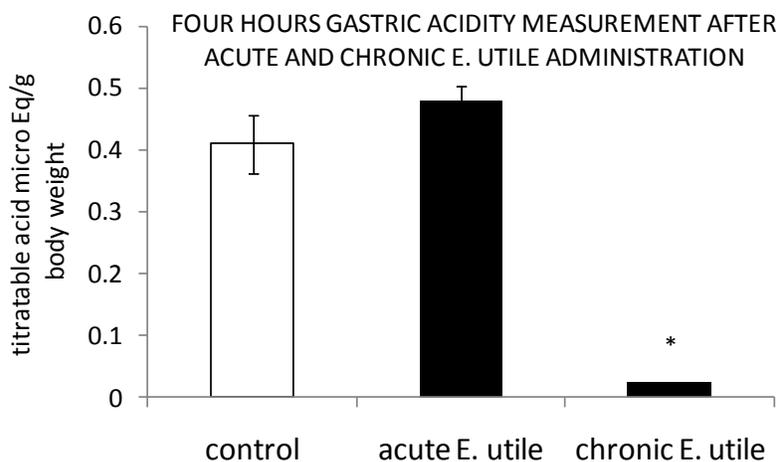


Fig. 3. Chronic administration of *E. utile* bark extract negatively affects gastric acid secretory mechanisms. The plot compares titratable acidity of 4h gastric effluents in rats given an acute dosage of 1ml of 100 mg ml⁻¹ extract of *E. utile* after ligation and in rats that were chronically fed with 100 mg ml⁻¹ aqueous extract of *E. utile ad libitum* over six weeks in their feeding bottles. The data indicates the extract inhibitory activity takes more than 4h to effect and the extract itself, having a pH range of 3 - 4.4, may contribute to titratable acidity of gastric effluents in the acute dosage rats. Chronic administration of *E. utile* extract resulted in block of gastric acid production using the 4h ligation period.

Effect of *E. utile* extract on peptic activity measurements in the Shay rats

The colour development after application of Ohnishi and Barr procedure (1978) to effluents in some of the animals treated with the extract was

reddish-purple instead of the bluish-purple obtained in the controls. In these specimens the optical density could not be read. Figure 1B shows the mean peptic activity for controls, ranitidine, and *E. utile* extract treated rats,

determined spectrophotometrically using a standard curve (Prino *et al.*, 1971 method). Control peptic activity (mg/dL bovine serum albumin digested) was 38.75 ± 4.05 . The extract produced a statistically significant fall in peptic activity, giving PA of 14.8 ± 4.67 ($p < 0.01$) comparable with ranitidine giving PA value of 3.4 ± 0.72 ($p < 0.001$).

Effects of *E.utile* extract on gastrointestinal muscle histamine receptors

The aqueous bark extract of *E.utile* did not produce contraction or relaxation of the guinea-pig ileum when given with increasing doses of $2-10 \times 10^{-3}$ g and observed with alternate doses of histamine (1-10-ug) that contracted the ileum in a dose-dependent manner. In the three point assay of (the extract of) *E.utile* against constant submaximal contractions induced by 10ug histamine, the extract of *E.utile* produced an antihistaminergic effect. In the latter study using atropinized bathing solution, *E.utile* given in doses of $10-160 \times 10^{-3}$ g antagonized 10 ug histamine-induced contractions by 28-62% in a dose-dependent manner (Figure 4). Mepyramine, but not ranitidine, also antagonized the contractile effect of histamine. Histamine gave a graded dose-response relationship (Figure 5), which was also established respectively in the presence of constant

doses of mepyramine or the extract (Figure 5). A plot of response (contraction amplitude) against log dose of histamine was shifted to the right by either mepyramine or the extract, both showing histamine antagonism. Automatic linear trendlines generated by Microsoft Excel 2007 are included in Figure 5 and show that mepyramine gave a parallel shift of the histamine dose-response curve typical of a competitive antagonism. Similarly the extract graph trendline indicated a non-parallel shift with a lowering of maximal response typical of a non-competitive antagonism (Figure 5).

DISCUSSION

In the 4h ligation rats the mean titratable gastric acidity was 0.38 uEq g^{-1} as compared to the controls of 0.53 uEq g^{-1} , the extract seeming not to have any antisecretory effect. Because the pH of the extract (3 – 4.4) was in the acid range, we thought that the ratio of extract: to gastric secretions might be important to be able to observe any antisecretory effect. Therefore we used increasing ligation periods. The extract significantly decreased titratable acidity of gastric effluents for the 6h ligation period, $p < 0.01$ (Figure 2).

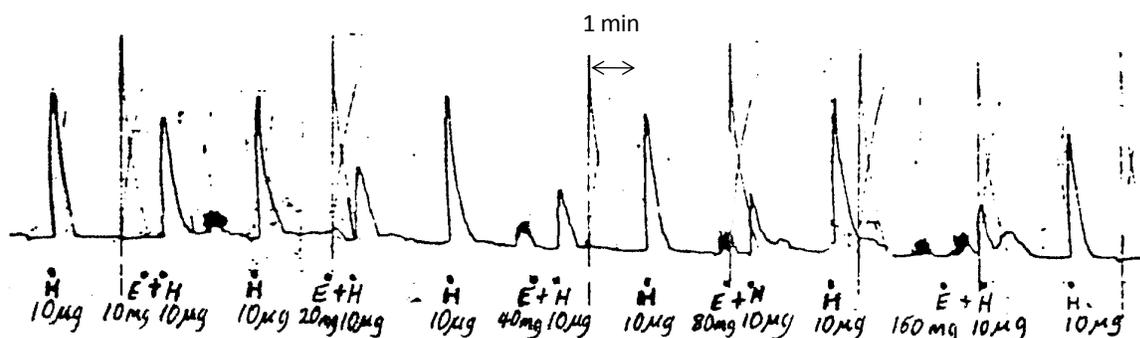


Figure 4.

E. utile bark extract antagonizes histaminergic contractions of the guinea-pig ileum in a dose-dependent manner. The ileum was bathed with atropinized Tyrode solution. The agonist, 10 ug histamine, which produced a large submaximal response was used against increasing doses of the extract added to the organ bath 30s before histamine. *E.utile* antagonized histamine-induced contractions in a dose-dependent manner.

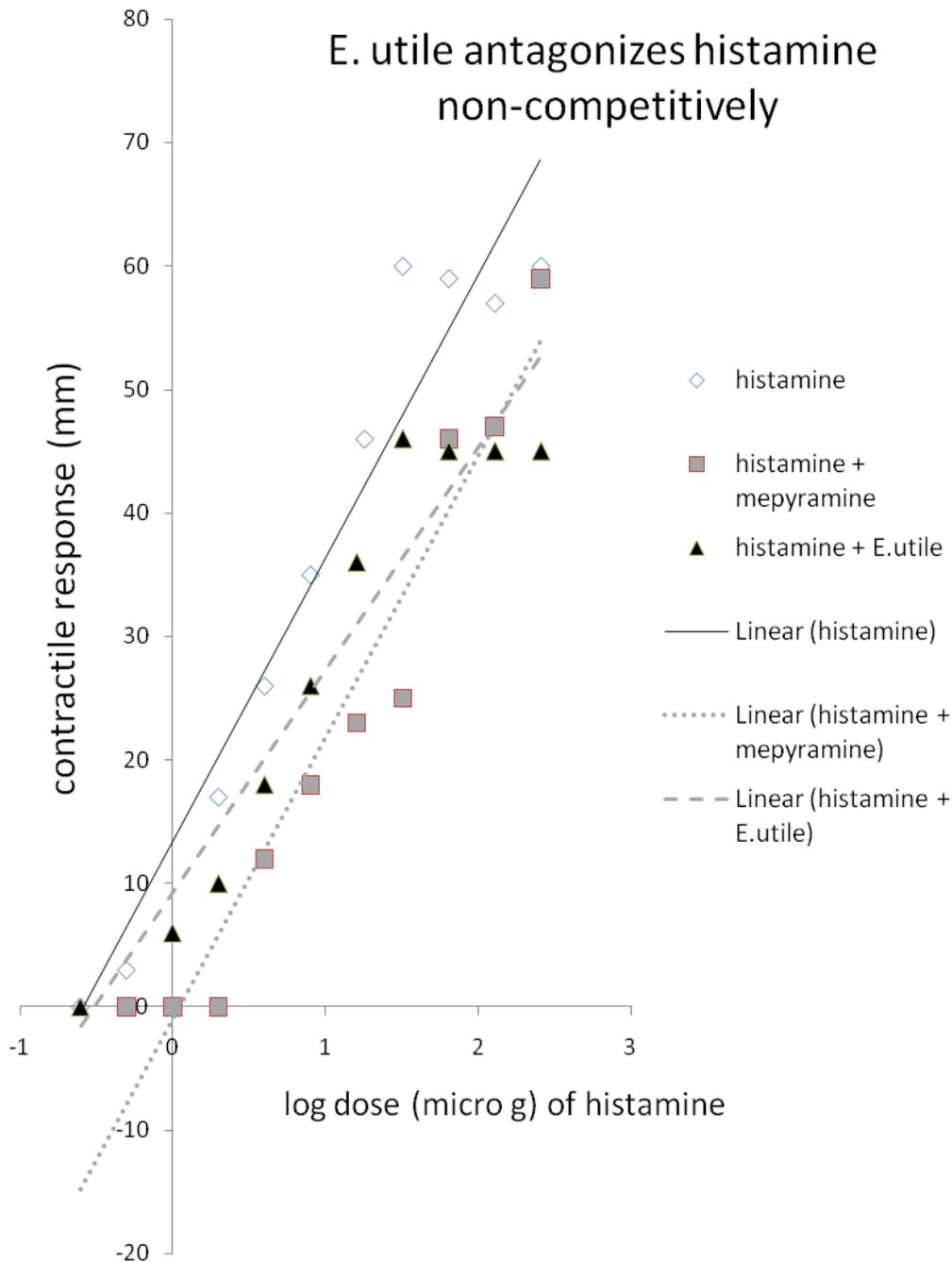


Figure 5.

E. utile bark extract antagonizes histaminergic contractions of the guinea-pig ileum non-competitively. The data are from tissue mounted in atropinized Tyrode solution. Contractile responses of the ileum were obtained for increasing doses of histamine. The responses to the same increasing doses of histamine were repeated in the presence of a constant dose of 5×10^{-6} M mepyramine or a constant dose of 10×10^{-3} g *E. utile* extract. Linear trend lines were automatically generated by Microsoft Excel 2007. Mepyramine gave a parallel shift of the histamine dose-response graph to the right typical of a competitive antagonism. *E. utile* extract gave a non-parallel shift to the right with a lowering of maximal response typical of a non-competitive antagonism.

This corresponds with the observed decreased in peptic activity produced by the extract even with 4h ligation. It may appear that the peptic activity inhibitory effect of the extract is more immediate (within 4h) than the acid secretion inhibitory effect of the extract (within 6 h). However in a different study of the antisecretory effect of *E. utile* using a continuous perfusion method, gastric acidity began to fall within 5 min of initiation of extract infusion (John 1994). In the present report, when the extract was administered chronically before the measurement of gastric acid output, the 4h ligation gastric effluent collection showed an inhibitory effect of the extract on titratable acid secretion (Figure 3) possibly because the ratio (secreted gastric acid/aqueous extract content) was higher than for the acute studies. The mean 4h measurements fell to $0.25 \mu\text{Eq g}^{-1} \text{H}^+$ after 6 weeks which indicates that the extract disturbed the function of the secretory mechanism during chronic intake. The biochemical assay for peptic activity by Ohnishi and Barr procedure (1978) was not compatible with the *E. utile* extract therefore we have used the data from the Prino *et al.* (1971) method. There was a statistically significant decrease in peptic activity for acute *E. utile* administration prior to 4 h ligation gastric effluent collections even when the data did not show decreased titratable acid secretion. The data indicates that *E. utile* showed antisecretory properties for both acidity and peptic activity. The effect on acid secretion might have been masked for short ligation periods because the extract itself has a low pH in the acid range. The extract did not show intrinsic activity on the guinea-pig ileum but antagonized the contractile effect of histamine in a dose dependent manner. Compared to mepyramine antagonism of histamine, the effect of the extract was not competitive antagonism and may not be directly at the H_1 -histamine receptor. In the gut, contraction is mediated through H_1 -receptor agonists and acid secretion is mediated through H_2 -receptor agonists (Dale and Laidlaw, 1910; 1911; Black *et al.*, 1972), therefore, the extract may be blocking both types of histamine receptors through a signal transduction mechanism. In unpublished work, we observed that the extract lacked any effect on cholinergic receptors of the

gut. In a different study of the antisecretory effect of the aqueous extract of *E. utile* using the Ghosh and Schild rat preparation, we observed that the extract may be also acting on proton pumps in the gastric mucosa. Because of the plurality of mechanisms that may be affected by the extract – histamine H_1 , histamine H_2 , and the proton pump – in the gastric mucosa, we need to investigate the molecular mechanisms involved, particularly the second messenger systems and how they are linked at receptor levels. The present documentation indicates that the aqueous bark extract of *E. utile* could be further studied for its usefulness in controlling gastric acid secretion.

Conclusion

Short term administration of effective doses of *E. utile* extract decreases gastric secretion of acid and also decreases gastric peptic activity. Long term administration of *E. utile* extract appears to negatively affect the mechanisms involved in gastric acid secretion. One mechanism by which the components of the *E. utile* bark extract produce anti-ulcer effect appears to be by antagonism of histamine receptors. The two properties of the *E. utile* extract on acid and pepsin may be valuable for patients with compromised mucosa due to aspirin ingestion (Ivey, 1988) or other related conditions. *E. utile* bark extract is a potential adjunct therapy that can minimize aggravation of mucosal damage by acid-peptic autodigestion.

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