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Original Article

Anti-spasmodic and Gastroprotective Activities of *Harungana madagascariensis* Leaf: A Traditional Anti-diarrhoea Remedy

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ABSTRACT

Background: *Harungana madagascariensis* Lam. ex Poir. (Hypericaceae) leaf extract is used by some communities in the Niger Delta to treat diarrhoea, ulcers, and wounds. **Objectives:** This study investigated the antispasmodic, antimotility and gastroprotective properties of methanol:dichloromethane (1:1) extract of *H. madagascariensis* leaves (HME). **Materials and Methods:** The antispasmodic activity was evaluated *in vitro* using actions on contractions of guinea pig ileum and rabbit jejunum provoked by spasmogens. The actions of HME on gastrointestinal transit was assessed *in vivo* using normal defaecation and charcoal meal transit time tests in rodents. The actions of HME on gastric ulcers produced by ethanol and indomethacin were investigated. The HME was also subjected to phytochemical analysis and acute toxicity tests. **Results:** The HME suppressed contractions of isolated rabbit jejunum and guinea pig ileum elicited by histamine and acetylcholine. The extract elicited significant (P<0.05) reduction of normal defaecation (12.50 - 100%) and gastrointestinal propulsion of charcoal meal in mice (17.60 - 43.08%). Additionally, the extract significantly (P<0.05) prevented both ethanol- and indomethacin-induced stomach ulcers. An oral LD_{50} >5000 mg/kg in mice was obtained by an acute toxicity assay on HME. **Conclusion:** The findings showed that the leaf of *H. madagascariensis* has gastroprotective, antispasmodic, and antimotility properties.
Keywords: Charcoal meal test, Gastric lesions, Gastrointestinal motility, Guinea pig ileum, Rabbit jejunum, Tyrode solution.

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INTRODUCTION

Harungana madagascariensis (the 'Haronga') Lam. ex Poir. (Hypericaceae) is a tropical, much branched (bushy) tree usually about small to medium in size with height of 4 -7 m, but sometimes reaching 10 - 25 m. The morphology and other characteristics have been described.[1-5] It is commonly called orange milk tree or blood tree (English), or 'oturu' (Nsukka; Igbo). Various parts (stem, root, leaf, fruits, etc.) of *H. madagascariensis* are widely used in the ethnomedicine of several African countries to manage numerous ailments, such as diarrhoea; wounds, including cuts, fresh circumcision wounds, and ulcers; malaria; anaemia; helminthiasis; haemorrhage; jaundice; headache; fever; infections, including sore throat, gonorrhoea, boils, and trypanosomiasis; skin diseases; among others.[5-16] In the Niger Delta region of Nigeria, the leaves are used to manage diarrhoea and malaria.[16] Earlier scientific studies demonstrated and documented the antimicrobial,[17-23] antihepatotoxic,[17] hypoglycaemic,[24] and antityphoid[25] properties of the leaf extracts. Furthermore, antibacterial prenylated anthracene derivatives[26] and astilbin (a flavanone)[21] have been isolated from the leaves.

Due to the ethnomedicinal use of *H. madagascariensis* leaf in the management of diarrhoea and ulcers, this study investigated how the leaf extract affected gastric lesions and gastrointestinal motility.

MATERIALS AND METHODS

Animals

Animals deployed for the investigation were male and female adult UN-FERH:NS outbred strain of albino mice (19-22 g), Sprague Dawley rats (150-200 g), guinea pigs (350 – 400 g), and rabbits (1.2 – 1.5 kg), bred in the Laboratory Animal Facility of the Department of Pharmacology and Toxicology, University of Nigeria, Nsukka. The animals were fed on water and pelleted meal. The procedures deployed complied with ethical guidelines stipulated by the National Institute of Health Guide for Care and Use of Laboratory Animals (Pub No. 85-23, updated 1985) and National Health Research Ethics Committee (NHREC/05/01/2016A).

Preparation of Extract

H. madagascariensis gathered in May, from Orba in Udenu Local Government Area of Enugu State,

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Nigeria, was verified by Mr. Alfred Ozioko, a Taxonomist at International Centre for Ethnomedicine and Drug Development (InterCEDD), Nsukka where a voucher specimen was stored (specimen no. InterCEDD/061). The leaves were cleaned, allowed to dry in the shade for seven days, and then milled into a coarse powder (2.83 kg) which was macerated in methanol:dichloromethane (1:1) at 25±1°C for 48 h. The plant material was repetitively washed with fresh solvent to obtain a clear filtrate, which was concentrated in a rotary evaporator under reduced pressure at 40°C to obtain 421.03 g of the methanol:dichloromethane extract (HME; 14.88% w/w).

The phytochemical constituents of HME were determined using established methods.[27-29]

Acute toxicity tests

The acute toxicity and lethality (LD_{50}) of HME were determined as described by Lorke(1983),[30] with slight modifications. Mice randomly placed in three groups (n=3) were orally given different doses of HME (10, 100 or 1000 mg/kg) dissolved in 80% (v/v) propylene glycol, and observed for 24 h for signs of acute toxicity and death. As no mice died, 1600, 2900 or 5000 mg/kg of HME were administered to a new set of animals at one dose per animal (n=1); they were observed for 24 h for signs of toxicity and death.

PHARMACOLOGICAL STUDIES

In vitro Pharmacological Studies

Studies on isolated guinea pig ileum

This was done as previously described.[31,32] Guinea pig sacrificed by cervical dislocation was exsanguinated, and a piece of the ileum was removed after discarding the part closest to the ileocecal joint. The ileal strip (approximately 2 cm in length) was mounted vertically under resting tension of 0.5 g in a 50 ml organ bath. The tissue was bathed in physiological salt solution (PSS) - Tyrode solution [(g/L); NaCl (8.0), KCl (0.2), $CaCl_2$ (0.2), $NaHCO_3$ (1.0), $MgCl_2$ (1.0), NaH_2PO_4 (0.5), glucose (1.0)], maintained at 37°C and aerated with air. The tissue was allowed to equilibrate for 60 min during which the physiological solution was changed every 10 min. Responses of the isolated ileum to graded concentrations of HME, acetylcholine (ACh) and histamine were recorded isometrically using an Ugo Basile Unirecorder (7050) through Ugo Basile isometric transducer (7004). Drug-tissue contact time was 1 min, and a 3 min time cycle was maintained. Also, the effects of HME on ACh (0.64 µg/ml)- and histamine (0.64 µg/ml)-induced contractions of the guinea pig ileum were recorded. The HME was added to the tissue bath and allowed to act for 3 min; subsequently, the standard agonist was added and allowed to act for 1 min before washing off. The experiments were carried out three times with separate guinea pigs.

Studies on isolated rabbit jejunum

This was done as previously described.[31,32] Rabbit sacrificed by cervical dislocation was exsanguinated, and a piece of the jejunum removed and freed of the mesentery. A jejunal strip (about 2 cm long) was mounted vertically in an organ bath (50 ml) under a resting tension of 0.5 g. The tissue was bathed in Tyrode solution [(g/L); NaCl (8.0), KCl (0.2), $CaCl_2$ (0.2), $NaHCO_3$ (1.0), $MgCl_2$ (1.0), NaH_2PO_4 (0.5), glucose (1.0)], maintained at 37°C and aerated with air. The tissue was allowed to equilibrate for 60 min with regular (every 10 min) changing of the PSS. Tissue responses to graded concentrations of HME, ACh and histamine were recorded isometrically using an Ugo Basile Unirecorder (7050) through Ugo Basile isometric transducer (7004). A 3-min time cycle was maintained, with a 1-min drug-tissue contact duration.

Additionally, the impact of HME on contractions of the rabbit jejunum provoked by ACh (1.28 µg/ml) and histamine (1.28 µg/ml) were recorded. After adding the HME to the tissue bath and giving it 3 min to act, the spasmogen was added and allowed to act for 1 min before washing off. The experiments were carried out three times with separate rabbits.

In vivo Pharmacological Studies

Normal defaecation test

The effect of HME on normal defaecation was investigated employing previously reported techniques,[33] with modifications[32] Rats subjected to 18 h fast and unlimited access to drinking water were randomly grouped (n = 5), and given 200, 400 or 800 mg/kg of HME through the oral route. Control groups received the vehicle 80% v/v propylene glycol (5 ml/kg), or loperamide (2 mg/kg) orally. The rats were housed singly in metal cages and faeces collected on white paper placed on a tray under each cage. Pulling out the tray allowed for observation of the faeces. The amount of faecal bolus each animal produced was measured every hour for 4 h.

Gastrointestinal motility test

The impact of HME on gastrointestinal motility was assessed by means of the charcoal meal test.[31,34] Mice subjected to 24 h fast and unlimited access to drinking water were randomly grouped (n = 5), and given 200, 400 or 800 mg/kg of HME. Control groups received the vehicle 80% v/v propylene glycol (5 ml/kg), or atropine (10 mg/kg). Thirty minutes later, charcoal meal (5% activated charcoal suspended in 10% aqueous solution of tragacanth powder) was given to each animal. All treatment was done through the oral route. Thirty minutes after administering the charcoal meal, the animals were sacrificed in a chloroform chamber and dissected to carefully identify and ligate the small intestine at both the pyloric sphincter and where the charcoal meal stopped (to prevent interfering with the charcoal meal whilst handling). The distance traversed by the charcoal meal from the pylorus, and the distance from the pylorus to the ileocecal junction (small intestine's length) were measured. The degree of intestinal propulsion (%) of the charcoal meal was determined by applying the equation:[32]

$$IP\ (\%) = (DT/TL)\ 100$$

Where:

IP = Intestinal propulsion; DT = Distance traversed by the charcoal meal; TL = Total length of the small intestine

Inhibition (%) of propulsion was computed in proportion to the control by applying the equation:[32]

$$\text{Inhibition of propulsion } (\%) = 100[1-(a/b)]$$

Where: a = IP of treated animals; b = IP of control animals

Indomethacin-induced ulcer

Stomach ulcers were produced by applying earlier reported techniques,[35] with slight modifications.[32] Rats subjected to 24 h fast were randomly distributed among five groups (n = 5), and given 200, 400 or 800 mg/kg of HME. Control animals received 5 ml/kg of the vehicle (80% v/v propylene glycol) or ranitidine (100 mg/kg). All treatment was done through the oral route. After 30 min, animals were given indomethacin 40 mg/kg, and sacrificed 8 h later in a chloroform chamber. The stomachs were removed, opened along the greater curvature and rinsed under a stream of water. Erosions formed on the glandular portion of the stomach were observed and each graded using a scale of 0-3 based on the length of the ulcer; 0 = normal; 1 = <1mm; 2 = 1- 2 mm; 3 = >2 mm.[36] Mean ulcer

score for each group was calculated and expressed as the Ulcer Index (UI). Ulcer protection (%) was computed with the following equation:

$$\text{Ulcer protection (\%)} = 100[1 - y / z]$$

Where:

y = Ulcer index of treated group

z = Ulcer index of control group[32]

Absolute ethanol-induced ulcer

Gastric ulceration was induced as described by Robert (1979).[37] Rats subjected to 24 h fast, were randomly distributed among five groups (n = 5), and given 200, 400 or 800 mg/kg of HME. Control animals received 5 ml/kg of the vehicle (80% v/v propylene glycol) or ranitidine (100 mg/kg). Thirty minutes later, ulcer was produced in each animal by administering absolute ethanol (1 ml). All treatment was delivered through the oral route. An hour later, the animals were sacrificed in a chloroform chamber and the abdomen cut open. The stomach of each animal was detached, opened along the greater curvature, rinsed under a stream of water and observed for ulcers. Gastric lesions formed on the glandular portion of the stomach were observed and each given severity rating on a 0-7 scale based on the ulcers,[32] with some modifications. Where 0 = no ulcer, 1= one ulcer of length ≤ 0.5 cm; 2 = more than one grade 1 ulcer, 3 = One ulcer of length >0.5 cm but < 1 cm, 4 = more than one grade 3 ulcer, 5 = one ulcer of length ≥ 1 cm, 6 = more than one grade 5 ulcer, 7 = complete haemorrhagic lesion of the gastric mucosa. Mean ulcer score for each group was calculated and expressed as the Ulcer Index (UI). Ulcer protection (%) was computed using the equation:

$$\text{Ulcer protection (\%)} = 100[1 - y/z]$$

Where:

y = Ulcer index of treated group

z = Ulcer index of control group[32]

Statistical Analysis

Data obtained were analysed using one-way ANOVA in GraphPad Prism 8.3.0 (GraphPad Software Inc., San Diego, CA) and subjected to Dunnett's multiple comparison test. Results were shown as mean±SEM, and differences between the means of treatment and control groups considered significant at p<0.001, p<0.01 or p<0.05 as applicable.

RESULTS

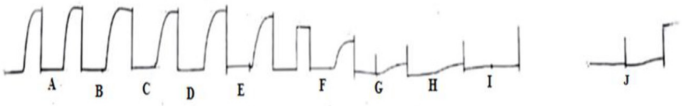
Phytochemical Analysis

The phytoconstituents detected in HME were alkaloids, tannins, saponins, reducing sugars, glycosides, flavonoids, carbohydrates, proteins, resins, terpenoids and steroids.

Effects of HME on contractions of the guinea pig ileum induced by standard spasmogens

The HME (0.04 - 5.12 mg/ml) produced a concentration-related inhibition of contractions of the guinea pig ileum induced by ACh (0.64 µg/ml); IC_{50} = 0.11 mg/ml (Figure 1, Figure 3, Table 1). Highest inhibition of 100±0.0% was elicited by 5.12 mg/ml (Figure 1, Figure 3).

The HME (0.04 - 0.32 mg/ml) caused a concentration-related inhibition of contractions of the guinea pig ileum induced by histamine (0.64 µg/ml); IC_{50} = 0.02 mg/ml (Figure 2, Figure 3, Table 1). Highest inhibition of 98.55±1.4% was elicited by 0.32 mg/ml (Figure 2, Figure 3).



A = ACh 0.64 µg/ml; B, C, D, E, F, G, H, I, = ACh (0.64 µg/ml) + HME 0.04, 0.08, 0.16, 0.32, 0.64, 1.28, 2.56, 5.12 mg/ml, respectively; J = ACh 40 µg/ml (re-established response)

Figure 1: Effect of *H. madagascariensis* leaf extract on acetylcholine-induced contractions of the guinea pig ileum.

A = Histamine 0.64 µg/ml; B, C, D, E = histamine (0.64 µg/ml) + HME 0.04, 0.08, 0.16, 0.32 µg/ml, respectively; F = histamine 40 µg/ml (re-established response)

Figure 2: Effect of *H. madagascariensis* leaf extract on histamine-induced contractions of the guinea pig ileum.

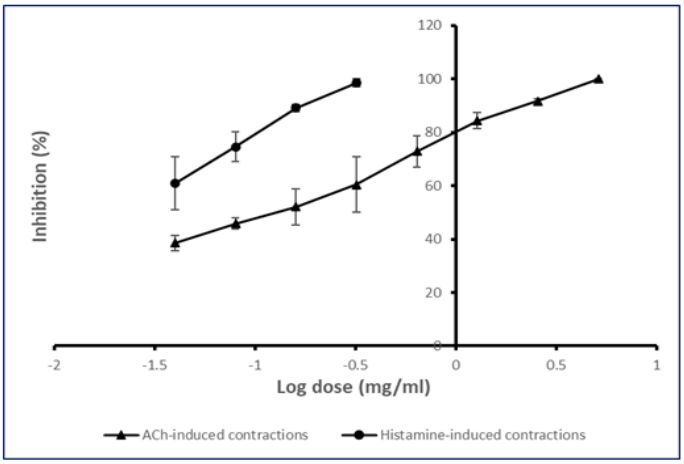


Figure 3: Inhibitory effects of *H. madagascariensis* leaf extract on spasmogen-induced contractions of the guinea pig ileum.

Table 1: Inhibitory effects of HME on spasmogen-induced contractions of intestinal tissues.

	Guinea pig ileum		Rabbit jejunum	
	Acetylcholine (0.64 µg/ml)	Histamine (0.64 µg/ml)	Acetylcholine (1.28 µg/ml)	Histamine (1.28 µg/ml)
IC_{50} (mg/ml)	0.11	0.02	1.40	0.03

IC_{50} = concentration that elicited 50% inhibition

Effects of HME on contractions of the rabbit jejunum induced by standard spasmogens

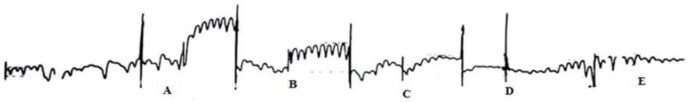
The HME had no perceptible effect on contractions of the rabbit jejunum, though it inhibited contractions of the jejunum produced by ACh (1.28 µg/ml) and histamine (1.28 µg/ml).

Increasing doses of HME (0.04 - 10.24 mg/ml) progressively inhibited contractions of the rabbit jejunum induced by ACh (1.28 µg/ml); IC_{50} = 1.40 mg/ml (Figure 4, Figure 6, Table 1). Highest inhibition of 95.69±0.5% was produced by 10.24 mg/ml (Figure 4, Figure 6).

Increasing doses of HME (0.04 - 0.32 mg/ml) progressively inhibited contractions of the rabbit jejunum induced by histamine (1.28 µg/ml);

A = ACh 1.28 µg/ml; B, C, D, E, F, G, H, I, J = ACh (1.28 µg/ml) + HME 0.04, 0.08, 0.16, 0.32, 0.64, 1.28, 2.56, 5.12, 10.24 mg/ml; K = ACh 40 µg/ml (re-established response)

Figure 4: Effect of *H. madagascariensis* leaf extract on acetylcholine-induced contractions of the rabbit jejunum.



A = Histamine 1.28 µg/ml; B, C, D, E = histamine (1.28 µg/ml) + HME 0.04, 0.08, 0.16, 0.32 mg/ml, respectively

Figure 5: Effect of *H. madagascariensis* leaf extract on histamine-induced contractions of the rabbit jejunum.

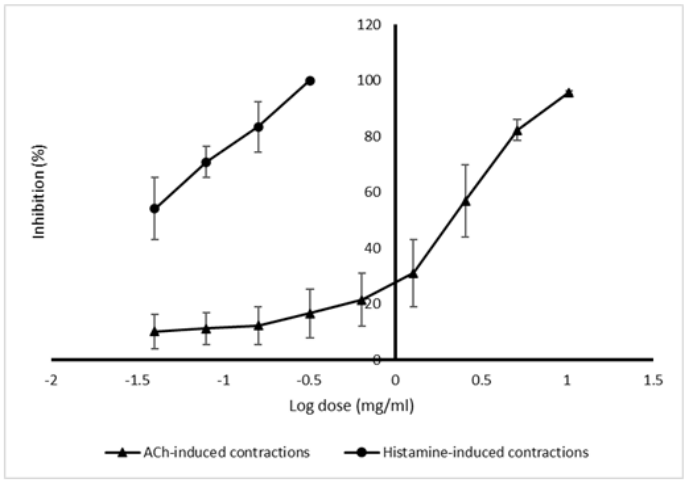


Figure 6: Inhibitory effects of *H. madagascariensis* leaf extract on spasmogen-induced contractions of the rabbit jejunum.

IC_{50} = 0.03 mg/ml (Figure 5, Figure 6, Table 1). Highest inhibition of 100±0.0% was elicited by 0.32 mg/ml (Figure 5, Figure 6).

Effect of HME on normal defaecation

The HME elicited significant and dose-related inhibition of normal defaecation, with 800 mg/kg producing 100% inhibition at 4 h (Table 2).

Effect of HME on gastrointestinal motility

The HME produced significant (P<0.05) and dose-related inhibition of gastrointestinal motility relative to control, as shown by decreased gastrointestinal propulsion and distance traversed by charcoal meal in HME-treated rats (Table 3).

Effect of HME on indomethacin-induced gastric lesions

The HME-treated rats had reduced indomethacin-induced gastric lesions, as shown by lower ulcer index values compared to control rats (Table 4, Figure 7). The HME produced significant and dose-related ulcer protection, though the effect of 400 mg/kg was slightly greater than that of 800 mg/kg (Table 4, Figure 7).

Effect of HME on absolute ethanol-induced gastric lesions

The HME-treated rats had less ethanol-induced gastric lesions, as shown by lower ulcer index values, compared to control rats (Table 5, Figure 8).

Table 2: Effect of HME on normal defaecation.

Treatment	Dose (mg/kg)	Mean no. of faecal boli			
		1h	2 h	3 h	4 h
HME	200	1.40±0.2*** (53.33)	1.40±0.2 (12.50)	1.00±0.5 (44.44)	0.80±0.2 (42.86)
	400	1.40±0.2*** (53.33)	1.00±0.3 (37.50)	1.00±0.3 (44.44)	0.40±0.2* (71.43)
	800	1.00±0.0*** (66.67)	0.40±0.2** (75.00)	0.60±0.2** (66.67)	0.00±0.0*** (100.00)
Loperamide	2	0.00±0.0*** (100.00)	0.00±0.0*** (100.00)	0.00±0.0*** (100.00)	0.00±0.0*** (100.00)
Control	-	3.00±0.5	1.60±0.2	1.80±0.4	1.40±0.2

n=5; *P<0.05, **P<0.01, ***P<0.001 compared to control (one-way ANOVA; Dunnett's post hoc test); HME = methanol:dichloromethane (1:1) extract of *H. madagascariensis* leaf; values in parentheses represent inhibition of normal defaecation relative to control

Table 3: Effect of HME on gastrointestinal motility.

Treatment	Dose (mg/kg)	Total length of intestine (cm)	Distance traversed by charcoal meal (cm)	Intestinal propulsion (%)	Inhibition of propulsion (%)
HME	200	47.68 ± 1.4	31.08 ± 0.3	65.35 ± 1.4*	17.60
	400	50.84 ± 0.3	27.04 ± 0.7	53.21 ± 1.5*	32.91
	800	49.30 ± 0.9	22.26 ± 0.8	45.14 ± 1.4*	43.08
Atropine	10	47.60 ± 0.6	15.58 ± 1.2	32.79 ± 2.6*	58.66
Control	-	45.60 ± 1.8	36.00 ± 0.8	79.31 ± 2.7	-

n=5; *P<0.05, compared to control (one-way ANOVA; Dunnett's post hoc test); HME = methanol:dichloromethane (1:1) extract of *H. madagascariensis* leaf; inhibition of propulsion (%) was calculated relative to control

Table 4: Effect of HME on indomethacin-induced gastric lesions.

Treatment	Dose (mg/kg)	Ulcer index	Ulcer Protection (%)
HME	200	10.50±3.0	15.32
	400	2.80 ±1.4*	77.42
	800	3.40 ± 0.7*	72.58
Ranitidine	100	1.40±0.9*	88.71
Control	-	12.40±5.2	-

n=5; *P<0.05 compared to control, ANOVA Dunnett's *post hoc* test; HME = methanol:dichloromethane (1:1) extract of *H. madagascariensis* leaf; ulcer protection (%) was calculated relative to control.

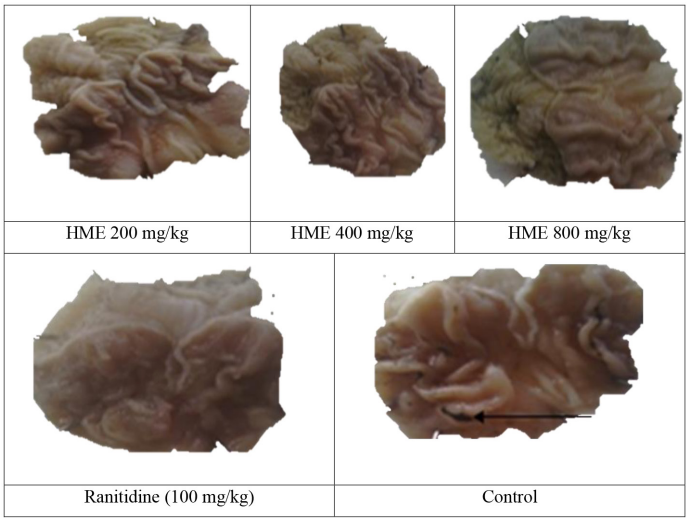


Figure 7: Effect of *H. madagascariensis* leaf extract on indomethacin-induced gastric ulcer in rats.

Table 5: Effect of HME on absolute ethanol-induced gastric lesions.

Treatment	Dose (mg/kg)	Ulcer index	Ulcer Protection (%)
HME	200	3.25±0.8	53.57
	400	2.00±0.3*	71.43
	800	2.00±1.2*	71.43
Ranitidine	100	2.25±1.0*	68.86
Control	-	7.00±2.0	-

n=5; **P*<0.05 compared to control, ANOVA, Dunnett's *post hoc* test; HME = methanol:dichloromethane (1:1) extract of *H. madagascariensis* leaf; ulcer protection (%) was calculated relative to control.

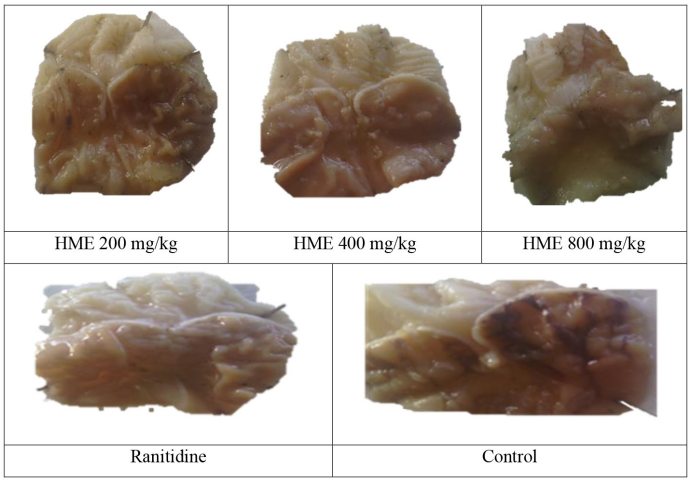


Figure 8: Effect of *H. madagascariensis* leaf extract on ethanol-induced gastric ulcer in rats

The HME produced significant and dose-related protection against gastric mucosal damage by absolute alcohol, though 400 and 800 mg/kg elicited equal degrees of protection (Table 5, Figure 8).

DISCUSSION

The findings of this study demonstrated that *H. madagascariensis* leaf extract inhibited contractions of intestinal tissues induced by ACh and histamine, inhibited normal defaecation and gastrointestinal (GI) propulsion, and protected against indomethacin- and ethanol-induced gastric ulcers.

The actions of the extract on spasmogen-induced contractions of isolated intestinal tissues were evaluated to provide insight into the purported use of the leaves to treat diarrhoea.[16] Results demonstrated ability of HME to attenuate contractions of the guinea pig ileum and rabbit jejunum induced by ACh and histamine, effects that are known to impede gut motility. Though HME on its own did not relax these intestinal tissues, inhibition of contractions induced by known endogenous spasmogens strongly indicates it may diminish the typical GI tract tone, and may principally be responsible for diminution of GI transit and normal defecation observed.

The HME caused significant ($P<0.05$) reduction of quantity of faecal bolus produced by normal rats from 1 - 4 h, suggesting inhibition of GI peristalsis and motility. Further investigation demonstrated the ability of the extract to inhibit GI motility, as shown by increase in charcoal meal transit duration *in vivo*. The main cause of the increased transit time is antimotility action which reduces GI propulsion and delays stomach emptying. Inhibition of small intestine's propulsion controls diarrhoea by averting rapid evacuation of GI contents.

Some excitatory and inhibitory neurotransmitters, including ACh, serotonin, vasoactive intestinal peptide, tachykinins (e.g., substance P), and nitric oxide control the motor and secretory processes of the GI tract.[38,39] Excitation-contraction coupling occurs in the GI smooth muscles as a result of an increase in intracellular Ca^{2+} concentration induced by ACh and other excitatory neurotransmitters.[39] The antimotility effect of *H. madagascariensis* leaf may, if not entirely, be due to non-specific action, as evidenced by HME's capacity to suppress spasmogen-induced contractions of the ileum and jejunum.

Diarrhea and other hyperactive gut diseases are likely to be assuaged by a substance that has the capacity to block the effects of excitatory neurotransmitter(s) or produce non-specific inhibitory action (e.g., antagonism of Ca^{2+}). During diarrhoea, notwithstanding the spasmogenic effects of luminal contents, antispasmodic action results in diminished motility and propulsion of GI contents. The decreased GI motility extends the luminal contents' stay in the gut, giving more time for water absorption and solidification of faeces.

The leaf extract, also utilized in ethnomedicine to treat ulcers, was further evaluated for antiulcer effects. Several orthodox medicines (e.g., muscarinic antagonists) and medicinal plants known to reduce GI motility have gastroprotective actions.[31] The extract produced significant and dose-related protection of the rat gastric mucosa in indomethacin- and ethanol-induced gastric ulcers.

Indomethacin causes gastroduodenal ulceration by inhibiting prostaglandins synthesis,[40] increasing gastric acid secretion,[40] and producing free radicals.[41] By inhibiting the production of prostaglandins, indomethacin and other NSAIDs enhance the predisposition to gastric mucosal lesions by preventing the vasodilator, antisecretory, and other defensive actions of prostaglandins. Therefore, the capability of the extract to avert ulcers caused by indomethacin points to elevation of prostaglandin level in the stomach mucosa and cytoprotection.

Ethanol causes extensive haemorrhagic erosions on the stomach mucosa,[34] due to mechanisms such as decreased secretion of bicarbonate and gastric wall mucus;[42] reduction of endogenous glutathione and

prostaglandin levels;[43] increase in the production and release of leukotriene C4;[44] and substantial oxygen free radical generation, which causes a rise in lipid peroxidation and cell damage.[45] Additionally, ethanol increases histamine release and calcium ion influx.[43] The observed antihistamine effect of HME on the isolated intestinal tissues may contribute to its capacity to prevent ethanol-induced ulcers. Therefore, the effect of the extract in ethanol-induced gastric lesions implies cytoprotective action that may be mediated by augmentation of mucosal defense mechanisms.

CONCLUSION

This study established that components of *H. madagascariensis* leaf exert antispasmodic, antimotility, and protective effects on the gastrointestinal system.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ACKNOWLEDGEMENT

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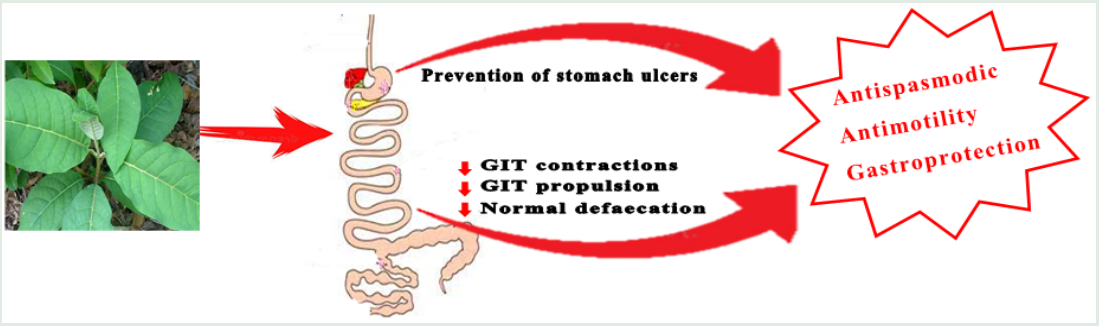
GRAPHICAL ABSTRACT



SUMMARY

Harungana madagascariensis Lam. ex Poir. (Hypericaceae) leaf is used by some communities in the Niger Delta, Nigeria to treat diarrhoea, ulcers, and wounds. This study investigated the antispasmodic, antimotility and gastroprotective properties of methanol: dichloromethane (1:1) extract of *H. madagascariensis* leaves. The results of this study established that H. madagascariensis leaf extract inhibited contractions of isolated rabbit jejunum and guinea pig ileum elicited by histamine and acetylcholine. The extract inhibited normal defaecation, and gastrointestinal propulsion of charcoal meal in mice. Additionally, the extract prevented both ethanol- and indomethacin-induced stomach ulcers.

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