Journal of Pharmacology and Toxicology

ISSN 1816-496X

Academic Journals Inc.

www.academicjournals.com

OPEN ACCESS Journal of Pharmacology and Toxicology

ISSN 1816-496X
DOI: 10.3923/jpt.2023.132.138

Research Article
Acute Toxicity Study, Anti-Secretory and Antiacid Activities of a Herbal Formulation on Induced Gastric Ulcer

[1]N'Guessan Jean-Baptiste Oussou, [1]Ehilé Hervé Ehile, [1]Kouakou Léandre Kouakou, [2]Mathieu Nitiema, [1]Kouamé Landry Yoboue, [1]Angoué Paul Yapo and [1]Ehouan Etienne Ehile

[1]Laboratory of Physiology, Pharmacology and Pharmacopoeia, Nangui Abrogoua University, Abidjan, Côte d'Ivoire
[2]Laboratory for the Research, Development of Phytomedicines and drugs, Institute for Research in Health Sciences, National Center for Scientific and Technological Research, Ouagadougou, Burkina Faso

Abstract

Background and Objective: Gastric ulcer is prevalent in Sub-Saharan African countries where people use medicinal plants for their treatment without knowing the probable side effects. This study was carried out to evaluate the acute toxicity study, the anti-secretory and the anti-acid activities of a herbal formulation made with *Macaranga barteri* and *Terminalia superba* (AEMbTs) on pylorus ligation-induced gastric ulcers in rats. **Materials and Methods:** The acute toxicity study using a single dose of 5000 mg kg^{-1} b.wt., of AEMbTs was conducted on 10 female rats following the Organisation for Economic Co-operation and Development (OECD) (420) guidelines. As for the anti-ulcer effects, 54 rats were divided into nine groups of six rats each. The rats in Group I (non-ulcerative rats) received distilled while the other groups were administered distilled water, Maalox, cimetidine and five doses of AEMbTs (31.25, 62.50, 125, 250 and 500 mg kg^{-1} b.wt.), respectively. As 1 hr later, the pylori of the rats were ligated. The rats were sacrificed, the stomach contents of each rat were collected, the pH and the gastric acidity were determined, then the ulcerations were measured and the inhibition percentages of the ulcerations were calculated 6 hrs later. **Results:** The AEMbTs didn't show any signs of toxicity. Hence, its 50% Lethal Dose (LD_{50}) was greater than 5000 mg kg^{-1} b.wt. The results, also, indicated that AEMbTs significantly reduced the ulcer surface areas, volume and gastric acidity while increasing the gastric pH of the rats compared to those with no treatment. **Conclusion:** Therefore, AEMbTs are safe and have anti-secretory and anti-acid activities on pylorus ligation-induced gastric ulcers in rats.

Key words: *Macaranga barteri*, *Terminalia superba*, toxicity, gastric ulcer, pylorus

Citation: Oussou, N.G.J.B., E.H. Ehile, K.L. Kouakou, M. Nitiema, K.L. Yoboue, A.P. Yapo and E.E. Ehile, 2023. Acute toxicity study, anti-secretory and antiacid activities of a herbal formulation on induced gastric ulcer. J. Pharmacol. Toxicol., 18: 132-138.

Corresponding Author: N'Guessan Jean-Baptiste Oussou, Laboratory of Physiology, Pharmacology and Pharmacopoeia, Nangui Abrogoua University, Abidjan, Côte d'Ivoire

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Gastric ulcer is a very common disease of the digestive system characterized by the appearance of more or less large lesions on the stomach wall[1]. These lesions which result from an imbalance between various aggression factors and the defense mechanisms of the gastric mucosa are under the influence of certain factors (spices, alcohol, tobacco), drugs (salicylates, non-steroidal anti-inflammatory drugs), psychogenic (stress) and infectious (*Helicobacter pylori*)[2,3]. Known worldwide, gastric ulcer affects less than 55% of people in Europe, while in Sub-Sahara African countries, it is estimated to be more than 75%[4]. In Côte d'Ivoire, an epidemiological study carried out in a hospital setting on peptic ulcers revealed a prevalence of 26.9%, in 2020[5]. To date, modern medicine offers a large number of synthetic drugs with antacids, antisecretory and cytoprotective effects for the treatment of gastric ulcer[6]. Despite the progress of this medicine, the use of plants remains an important source of care in some countries of the world. This use of medicinal plants is social, economic and cultural but above all accessible to populations for the treatment of chronic diseases such as gastric ulcers[7]. Moreover, it is very common to see that in the treatment of diseases, populations frequently resort to combinations of plants in the quest for greater efficiency. Most of the time, the traditional use of these plants has no consequences for the users' health. This is why the World Health Organization recommends verifying the safety and effectiveness of herbal formulations before their use[7]. It is with this in mind that two plants from the Southern Ivorian forest, known to have anti-ulcer properties[8] were used in this research work. Furthermore, Ehilé *et al.*[9] and Kouakou *et al.*[10] conducted studies on *Macaranga barteri* and *Terminalia superba*, respectively to assess their effect against an experimentally induced gastric ulcer in rats. The results of these two research works revealed cytoprotective, antisecretory and antacid properties. Thus, this work aims to determine the safety and evaluate the anti-secretory and anti-acid effects of this herbal formulation made with *Macaranga barteri* leaves and *Terminalia superba* stem bark (AEMbTs) on gastric ulcers induced by pyloric ligation in rats.

MATERIALS AND METHODS

This study was carried out from September, 2021 to March, 2022 and has been defended in August, 2023 by a Master's degree student.

Plant: The plant material consisted of the stem bark of *Terminalia superba* Engl. and Diels (Combretaceae) and fresh leaves of *Macaranga barteri* Müll.Arg (Euphorbiaceae). These samples were identified by botanists from Nangui Abrogoua University and authenticated using specimens kept in the National Floristic Center (CNF) of the Félix Houphouët-Boigny University (Abidjan, Côte d'Ivoire) under the voucher number 14735 of April 06, 1979, for *M. barteri* and 10477 of February 26, 1969, for *T. superba*.

Animals: The experiments were conducted on adult albino rats (*Rattus norvegicus*). The rats were housed in the same conditions as our previous works. The animals were provided by the animal house and kept in the animal house of the Laboratory of Physiology, Pharmacology and Pharmacopoeia (LPPP) of the Nangui Abrogoua University (Abidjan). The different experimental protocols were followed as the protocols for protecting experimental animals[11].

The acute toxicity study was conducted on female rats aged about 5 to 7 weeks, weighing 110 and 125 g. As for the gastric ulcers study, male and female rats aged between 12 to 16 weeks, with a body weight ranging between 180 and 200 g were used.

Chemicals and drugs: The chemicals and drugs used in this study were aluminum hydroxide (Maalox®, Sanofi Aventis, France), Cimetidine (Saint Louis, Missouri, United States of America), ether (VWR International Geldenaaksebaan 464-B-3001, Leuven, Belgium) and sodium hydroxide (Sigma, United States of America).

Methods

Preparation of the herbal formulation: *Terminalia superba* stem bark and *Macaranga barteri* leaves were harvested, cleaned, cut into small pieces and dried in a room at 25°C for 1 week. The aqueous extracts of the leaves of *M. barteri* and the stem barks of *T. superba* were separately prepared according to the methods described by Ehilé *et al.*[9] and Kouakou *et al.*[10], respectively. One hundred grams of each powder (leaves of *M. barteri* or bark of *T. superba*) was mixed with 1 L of distilled water. The mixture was boiled and thereafter filtered through absorbent cotton and a Whatman No.1 filter paper. The filtrate was dried at 45°C for 48 hrs using an oven (Friucell, Germany). The extracts of each plant were stored in a refrigerator at 8°C. The different doses of the herbal formulation were prepared extemporaneously by mixing the same amount of both extracts before dissolving them into distilled water.

Acute oral toxicity study: The acute oral toxicity study of AEMbTs was conducted according to the OECD guidelines 425[12]. It consisted of administering AEMbTs at doses of 5000 mg kg^{-1} b.wt., to a group of (10) female rats weighing between 110 and 125 g compared to another group of ten (10) female rats gavaged with distilled water (1 mL/100 b.wt.). The animals were observed for 14 days then the 50% Lethal dose (LD_{50}) was determined.

Induction of gastric lesions using the pyloric ligation model: The method was described by Hayase and Takeuchi[13]. This method was used to assess the antacid and/or antisecretory properties of AEMbTs. Fifty-four rats were fasted for 24 hrs and divided into nine groups of six rats each. The rats of Group I and II received by oral route distilled water (1 mL/100 g b.wt.). Those of Groups III and IV received orally cimetidine (12 mg kg^{-1} b.wt.) and Maalox (50 mg kg^{-1} b.wt.), respectively. As for the rats of Groups V to IX, they were orally administered 31.25, 62.5, 125, 250 and 500 mg kg^{-1} b.wt. of AEMbTs, respectively. One hour after the various administrations, incisions, under the last left lateral rib of rats, except those of Group I, was made under ether anesthesia. The pylori were, thereafter, tied off and the incisions were sutured. As 6 hrs after ligating the pylori, the animals are sacrificed by an overdose of ether. The stomach contents of the rats were collected and the volume was determined. The gastric pH was determined using a pH meter (HANNA HI 9025, United States of America). The stomach contents were centrifuged at 3000 rpm for 10 min. The 1 mL of the supernatant was collected in tubes and the gastric acidity was determined by titration with a sodium hydroxide solution (0.1N, NaOH). The gastric acidity was calculated using the following formula:

$$\text{Acidity (mEq L}^{-1}) = V(NaOH) \times \frac{N(NaOH)}{0.1} \times 100$$

With:
V(NaOH) = Volume of NaOH
N(NaOH) = Normality of NaOH

The gastric mucus was collected and weighed. The ulcers were classified according to the classification scale (score) of gastric lesions described by Zhang *et al.*[14].

As 0.5: Absence of ulcer (normal mucosa), 0.5: Dilation of blood vessels (presence of redness), 1: Small marks of ulcers, 1.5: Dilation of vessels blood and presence of ulcer marks, 2: Ulcers $\geq$3 mm long $\leq$5 mm long and 3: Ulcers >5 mm long.

Once the measurements were recorded, the ulceration index (UI) and the ulceration inhibition percentages (I%) were calculated using the formulas described by Nguelefack *et al.*[15]:

$$DUG = \frac{Score}{NUG}$$

With:
DUG = Degree of ulceration of the group
Score = Sum of points assigned to gastric lesions (score)
NUG = Number of ulcerative rats in the group

$$UI = DUG \times \frac{PUG}{100}$$

With:
UI = Group ulceration index
PUG = Percentage of ulcerated rats in the group

$$I(\%) = SU_t \times \frac{100}{SU_t - SU_e}$$

With:
I(%) = Ulceration inhibition percentage of the treated rats
Su_t = Ulceration surface of Group II (ulcerative non-treated rats)
SU_e = Ulceration surface of the treated groups

Statistical analysis: The data recorded during the different tests were classified and performed using GraphPad Prism 7.01 software (San Diego, California, United States of America). The results were expressed as Means±Standard error on the mean (m±SEM). The Student's t-test and the One-Way Analysis of Variance (ANOVA 1) were used to compare treated and control rats' batches. When significant differences were revealed, One-way ANOVA was completed by the Tukey-Kramer test as a *post hoc* test. The significance level was set at $p<0.05$.

RESULTS

Acute oral toxicity study of AEMbTs: The result of the acute oral toxicity study of the herbal formulation (AEMbTs) showed no motor and respiratory difficulties in the rats. No change in

fecal matter and no oral, nasal, or anal bleeding were observed. There were no changes in the general appearance of the rats (tremor, appearance of hairiness). The herbal formulation did not cause any mortality. This suggests that AEMbTs are not toxic at a higher and single dose with a 50% (LD_{50}) which is more significant than 5000 mg kg^{-1} b.wt.

Effects of AEMbTs on pyloric ligation-induced gastric injury

Effects of AEMbTs on macroscopic parameters: The effects of the different treatments on the gastric lesions induced by pyloric ligation of the rats were shown in Fig. 1 and Table 1. The results showed that the rats in Group I, (non-pyloric ligated rats) didn't show any gastric mucosa lesions. Therefore, the parameters such as the ulceration surface, the ulceration index and the score are all nil. On the other hand, the rats in Group II (pyloric ligated rats) without any pretreatment highlighted a significant increase ($p<0.001$) of these parameters compared to Group I. the values were 2.50±0.28, 30.8±5.16 and 2.96±0.40 mm^2 for the score, the ulceration area and the ulceration index, respectively. The mucus produced by the rats of Group II was significantly reduced ($p<0.001$) when compared to that of the rats of Group I (128.2±3.35 vs 42.5±3.23 mg).

The score, surface area and ulceration index of rats from Groups III and IV pretreated with cimetidine (12 mg kg^{-1} b.wt.) and Maalox (50 mg kg^{-1} b.wt.) decreased significantly

Table 1: Macroscopic parameters of gastric lesions induced by pyloric ligation

Group	US (mm^2)	UI	Score	I(%)	Mucus (mg)
Group I, non-ulcerative rats (distilled water)	00±00	00±00	00±00	-	128.2±3.35
Group II, ulcerative rats (distilled water)	30.8±5.16$^{###}$	2.96±0.40$^{###}$	2.50±0.28$^{###}$	-	42.5±3.23$^{##}$
Group III (Cimetidine 12 mg kg^{-1} b.wt.)	0.77±0.85***	0.25±0.15**	0.27±0.06***	98.54	89.00±6.52***
Group IV (Maalox 50 mg kg^{-1} b.wt.)	0.95±0.64***	0.32±0.16***	0.36±0.14***	98.19	96.00±5.26***
Group V (AEMbTs 31.25 mg kg^{-1} b.wt.)	15.3±1.8***	1.75±0.14*	1.63±0.23*	50.32	64.00±4.93**
Group VI (AEMbTs 62.5 mg kg^{-1} b.wt.)	11.5± 4.48***	1.13±0.23*	1.13±0.23***	62.66	80.3±1.75***
Group VII (AEMbTs 125 mg kg^{-1} b.wt.)	7.06±1.29***	1.00±0.2*	0.87±0.12***	77.07	90.8±7.89***
Group VIII (AEMbTs 250 mg kg^{-1} b.wt.)	3.25±0.47***	0.62±0.12***	0.42±0.12***	89.44	101.00±2.08***
Group IX (AEMbTs 500 mg kg^{-1} b.wt.)	0.75±0.85***	0.30±0.13***	0.22±0.06***	97.56	107.30±5.76***

$^{###}p<0.001$: Comparison between Group I (non-ulcerative rats) and those of Group II (ulcerative rats). **p<0.01, ***p<0.001: Significant difference between the values of the same column and those of Group II (ulcerative rats), US: Ulceration surface, UI: Ulcer index, I: Inhibition percentage, AEMbTs: Aqueous extract of *Macaranga barteri* leaves and trunk bark of *Terminalia superba* and n = 6 rats/group

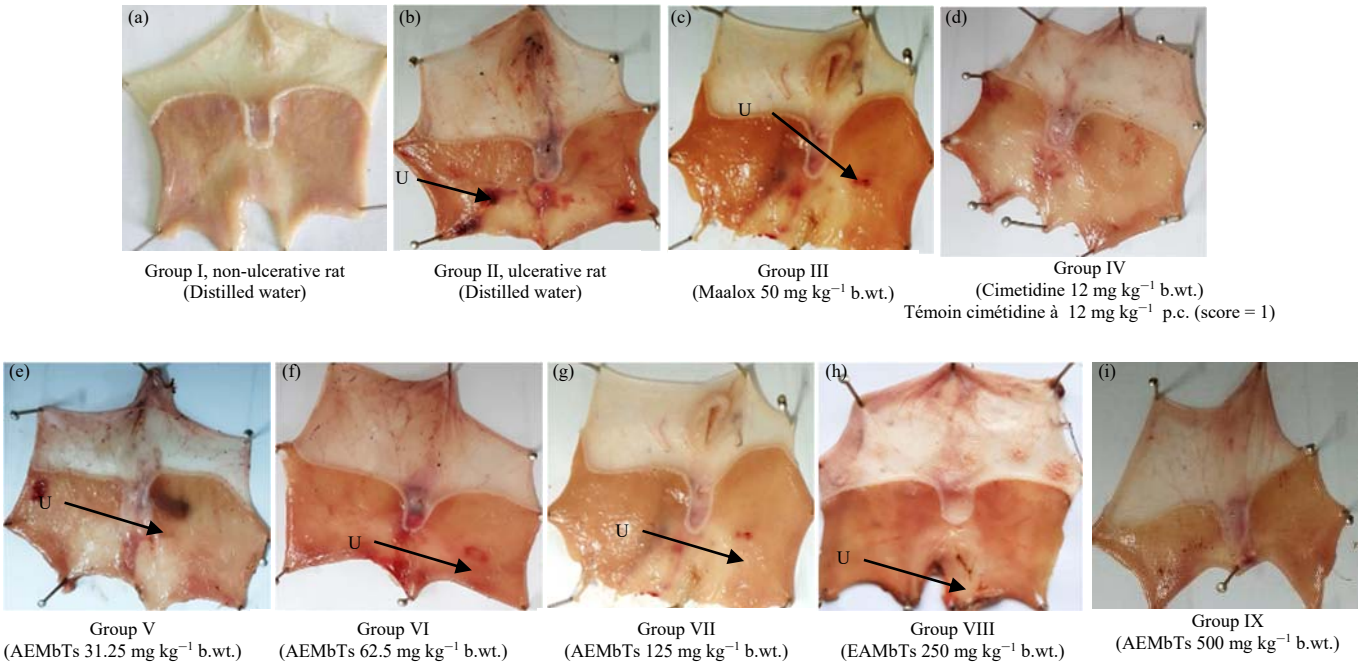

Fig. 1(a-i): Photographs of stomachs after different treatments in the pyloric ligation-induced rats

AEMbTs: Aqueous extract of *Macaranga barteri* leaves and *Terminalia superba* stem bark, U: ulceration

Table 2: Gastric secretions parameters after different treatments in the pyloric ligation-induced rats

Group	Gastric pH	Gastric volume (mL)	Gastric acidity (mEq L^{-1})
Group I, non-ulcerative rats (distilled water)	-	-	-
Group II, ulcerative rats (distilled water)	1.35±0.08	5.38±0.23	61.80±2.30
Group III (Cimetidine 12 mg kg^{-1} b.wt.)	5.040±0.11***	2.25±0.25***	33.50±0.95***
Group IV (Maalox 50 mg kg^{-1} b.wt.)	4.970±0.35***	2.38±0.23***	35.80±3.75***
Group V (AEMbTs 31.25 mg kg^{-1} b.wt.)	2.20±0.09**	3.43±0.25***	47.74±5.63***
Group VI (AEMbTs 62.5 mg kg^{-1} b.wt.)	2.97±0.07***	3.08±0.32**	42.32±1.93***
Group VII (AEMbTs 125 mg kg^{-1} b.wt.)	3.60±0.25***	2.98±0.11***	43.30±2.02**
Group VIII (AEMbTs 250 mg kg^{-1} b.wt.)	4.17±0.38***	2.43±0.21**	38.41±0.62***
Group IX (AEMbTs 500 mg kg^{-1} b.wt.)	4.95±0.17***	1.75±0.32***	34.28±1.77***

***p<0.001: Significant difference between the values of the same column and those of rats II (ulcerative rats), AEMbTs: Aqueous extract of *Macaranga barteri* leaves and *Terminalia superba* stem bark and n = 6 rats/group

(p<0.001) compared to control Group II. The ulceration surface was 0.77±0.85 mm^2 for Group III and 0.95±0.64 mm^2 for Group IV, corresponding to a percentage inhibition of 98.54 and 98.19%, respectively. The ulceration index is 0.25±0.15 and 0.32±0.16, respectively in the rats of Groups III and IV. The score was 0.27±0.06 (Group III) and 0.36±0.14 (Group IV).

The two reference drugs induced a significant increase (p<0.001) in the secreted mucus which is worth 89.00±6.52 mg (Group III) and 96.00±5.26 mg (Group IV).

As for the rats in Groups V to IX, pretreated with AEMbTs at doses between 31.25 and 500 mg kg^{-1} b.wt., the results showed a dose-dependent significant (p<0.001) inhibition of the gastric ulcerations and an increase of the mucus produced compared to the Group II. The weight of the mucus secreted varied from 64.00±4.93 (Group IV) to 107.30±5.76 mg (Group IX), while the non-pretreated group (Group II) produced 42.5±3.23 mg of it.

Effects of AEMbTs on gastric secretions following different treatments: The effects of AEMbTs on the gastric secretions following pyloric ligation in rats were shown in Table 2. The results showed a gastric volume of 5.38±0.23 mL, a gastric pH of 1.35±0.08 and a gastric acidity of 61.80±2.30 mEq L^{-1} in ulcerative non-pretreated rats (Group II). The pretreatment of rats of Groups III and IV, respectively with cimetidine (12 mg kg^{-1} b.wt.) and Maalox (50 mg kg^{-1} b.wt.) and those of Groups IV to VI pretreated with AEMbTs at doses between 31.25 and 500 mg kg^{-1} b.wt. significantly (p<0.001) reduced the volume of gastric contents produced in rats following pyloric ligation compared to Group II rats. This volume varies in the rats pretreated with AEMbTs, from 3.43±0.25 mL (Group V) to 1.75±0.32 mL (Group IX). The gastric pH recorded following pyloric ligation increased significantly (p<0.05 and p<0.001) in rats pretreated with AEMbTs and ranges between 2.20±0.09 (Group V) and 4.95±0.17 Group IX. The gastric pH also increased significantly (p<0.001) in rats of Groups III and IV which received, respectively cimetidine and Maalox with respective values of 5.04±0.11 and 4.97±0.35. As for the gastric acidity, which was 61.80±2.30 mEq L^{-1} in ulcerative non-retreated rats (Group II) drops significantly (p<0.001) to 34.28±1.77 mEq L^{-1} in rats pretreated with AEMbTs (500 mg kg^{-1} b.wt.). Those of the rats of Groups III and IV, which received, respectively cimetidine and Maalox revealed respective values of 33.50±0.95 and 35.80±3.75 mEq L^{-1}.

The herbal formulation (AEMbTs) has antisecretory and anti-acid activities as it reduces gastric volume and acidity while increasing gastric pH. The effects of AEMbTs are close to those of cimetidine and Maalox on the gastric mucosa.

DISCUSSION

The results of the acute toxicity study of AEMbTs in rats showed that the extract did not cause any mortality at the doses of 5000 mg kg^{-1} b.wt. Therefore, the 50% Lethal dose (LD_{50}) exceeds 5000 mg kg^{-1} b.wt. The extract is not toxic at a higher and single amount in the short term. According to the Globally Harmonized Classification System (GHS) of chemical substances, the extract can be classified in category 5[16]. This result was justified by the conclusions of previous studies carried out on the toxicity of each of the two species of the formulation, which showed that the aqueous extract of *Macaranga barteri* leaves administered orally to rats is not toxic at a dose of 5000 mg kg^{-1} b.wt.[9]. On the other hand, Kouakou *et al.*[10] also concluded from their work that 5000 mg kg^{-1} b.wt. of the aqueous extract of the stem bark of *Terminalia superba* is non-toxic. The combination of these two extracts retains their harmlessness.

Regarding the effect of AEMbTs on gastric ulcers, the results revealed that this herbal formulation has real anti-ulcerogenic potential. This extract exerts anti-secretory and anti-acid activities against gastric lesions induced by rats' pyloric ligation. The results showed that AEMbTs significantly reduce the gastric lesions while promoting, at the same time,

a significant reduction in volume and gastric acidity by increasing the pH of gastric juice compared to the ulcerative rats. Indeed, ligating the pylorus leads to a rupture of the barrier of the gastric mucosa. This rupture occurred due to the overproduction of acid by the stomach or the reduction of protective factors such as mucus, bicarbonate and prostaglandin produced by the gastric mucosa[17]. This could explain the large areas of ulceration observed in ulcerative control rats that have not been pretreated. However, the results of this study showed that AEMbTs caused a significant reduction in gastric volume, free acidity, ulcer surface area and an increase in pH in a dose-dependent manner. This result indicated that active compounds such as tannins, flavonoids and saponins highlighted by the works of Ehilé *et al*.[9] and Kouakou *et al*.[10] could be responsible for the regulation of the action of the stomach acid mediators. Indeed, tannins are endowed with anti-ulcerogenic properties.

They neutralize ulcer progression through their protein precipitation effects. Their astringent action helps precipitation microproteins at the ulcer site, thus forming an impermeable layer on the mucosa, which prevents gastric secretions and protects the underlying mucosa from toxins and other irritants[18]. As for saponins, they protect the gastric mucosa against acidic effects by promoting the production of mucus[19]. These results indicated that the extract has antacid and antisecretory activity in pyloric ligation-induced gastric ulcer rats. These results are similar to those obtained by Tyagi *et al*.[20], who showed that aqueous extracts of *Aegle marmelos* and *Ocimum sanctum*, also containing tannins and flavonoids, reduce the acidity of gastric contents. The results of this study imply that AEMbTs are a good candidate for the treatment of gastric ulcers in view of their pharmacological effects. The herbal formulation can be recommended in case of ulcers linked to hypersecretion of acid in the stomach. This study has also shown that AEMbTs can be taken in high doses without any adverse effects. The study should be extended to other factors of gastric ulcers, mainly on the bacterium responsible for gastric ulcers (*Helicobacter pylori*).

CONCLUSION

The herbal formulation made with *Macaranga barteri* leaves and *Terminalia superba* stem bark (AEMbTs) is not toxic at higher and single doses with an LD_{50} more fabulous than 5000 mg kg^{-1} b.wt. The extract is endowed with anti-secretory and anti-acid activities in pyloric ligation-induced ulcers in rats' models by increasing the gastric pH and decreasing the gastric volume and acidity in pretreated rats. The mixture also inhibits gastric ulcers. The results of this research work could justify using these two herbs in traditional medicine to treat gastric disorders.

SIGNIFICANCE STATEMENT

The plants used in this recipe are used in traditional medicine by populations for their care. Research work undertaken in our laboratory has shown anti-ulcer effects of these. The aim of this work was to demonstrate the scientific evidence for the use of plant extracts in the treatment of ulcers by evaluating the acute toxicity, anti-secretory and anti-acid activities of a formulation based on *Macaranga barteri* leaves and *Terminalia superba* stem bark (AEMbTs) in gastric ulcers induced by pyloric ligation in rats. The results showed that AEMbTs are safe and effectively treat gastric ulcers through their antisecretory and antacid effects. The subsequent study to be carried out remains to verify the effect of the recipe on *Helicobacter pylori*, the bacterium responsible for gastric ulcers.

ACKNOWLEDGMENTS

The authors would like to be thankful to all other members of the Laboratory of Physiology, Pharmacology and Pharmacopoeia and the Laboratory for the Research-Development of Phytomedicines and Drugs for their encouragement and assistance during these investigations.

REFERENCES

1. Ramakrishnan, K., Frcse and R.C. Salinas, 2007. Peptic ulcer disease. Am. Fam. Physician, 76: 1005-1012.
2. Freston, J.W., 1988. The pathophysiological and pharmacological basis of peptic ulcer therapy. Toxicol. Pathol., 16: 260-266.
3. Kaunitz, J.D. and Y. Akiba, 2004. Gastroduodenal mucosal defense: Role of endogenous mediators. Curr. Opin. Gastroenterol., 20: 526-532.
4. Leclerc, H., 2006. Epidemiological aspects of *Helicobacter pylori* infection. Bull. Acad. Natl. Med., 190: 949-962.
5. Stanislas, D.A., K.G. Dimitri, B.A. Demba, Y. Abdélatif and M. Mamonman *et al*., 2020. Prevalence of gastroduodenal ulcers and gastric precancerous lesions in chronic gastritis due to helicobacter pylori according to the Sydney system: About 52 cases. Health Sci. Dis., 21: 16-20.
6. Sharifi-Rad, M., P.V.T. Fokou, F. Sharopov, M. Martorell and A.O. Ademiluyi *et al*., 2018. Antiulcer agents: From plant extracts to phytochemicals in healing promotion. Molecules, Vol. 23. 10.3390/molecules23071751.

7. O.M.S., 2013. WHO Strategy for Traditional Medicine 2014-2023 (In French). World Health Organization, Geneve, Switzerland, ISBN: 9789242506099, Pages: 75.
8. Oliver-Bever, B., 1986. Medicinal Plants in Tropical West Africa. Cambridge University Press, Cambridge, England, ISBN: 9780521268158, Pages: 375.
9. Herve, E.E., G.N. Bernard, K.K. Leandre, Y.A. Paul and E.E. Etienne, 2018. Acute toxicity and gastric anti-ulcer activity of an aqueous extract of the leaves of *Macaranga barteri* Müll.Arg (Euphorbiaceae) on rat models. J. Med. Plants Res., 12: 96-105.
10. Leandre, K.K., G.N. Bernard, B.N. Mathieu, K.B. Andre and A.K. Augustin *et al.*, 2013. Acute toxicity and anti-ulcerogenic activity of an aqueous extract from the stem bark of *Terminalia superba* Engl. and Diels (Combretaceae). World J. Pharm. Sci., 1: 117-129.
11. Ferdowsian, H.R. and N. Beck, 2011. Ethical and scientific considerations regarding animal testing and research. PLoS ONE, Vol. 6. 10.1371/journal.pone.0024059.
12. OECD, 2022. OECD Guidelines for the Testing of Chemicals, Section 4. In: Test No. 425: Acute Oral Toxicity: Up-and-Down Procedure, OECD (Ed.), OECD, Washington, pp: 1-28.
13. Hayase, M. and K. Takeuchi, 1986. Gastric acid secretion and lesion formation in rats under water-immersion stress. Dig. Dis. Sci., 31: 166-171.
14. Zhang, Y., J. Jia, Y. Ding, Y. Ma and P. Shang *et al.*, 2016. Alpha-boswellic acid protects against ethanol-induced gastric injury in rats: Involvement of nuclear factor erythroid-2-related factor 2/heme oxygenase-1 pathway. J. Pharm. Pharmacol., 68: 514-522.
15. Nguelefack, T.B., P. Watcho, S.L. Wansi, N.M. Mbonuh, D. Ngamga, P. Tane and A. Kamanyi, 2005. The antiulcer effects of the methanol extract of the leaves of *Aspilia africana* (Asteraceae) in rats. Afr. J. Tradit. Complementary Altern. Med., 2: 233-237.
16. UNECE, 2021. Globally Harmonized System of Classification and Labeling of Chemicals [In French]. 9th Edn., United Nations, New York, ISBN: 9789210052146, Pages: 598.
17. Dhuley, J.N., 1999. Protective effect of *Rhinax*, a herbal formulation against physical and chemical factors induced gastric and duodenal ulcers in rats. Indian J. Pharmacol., 31: 128-132.
18. de Jesus, N.Z.T., H. de Souza Falcão, I.F. Gomes, T.J. de Almeida Leite and G.R. de Morais Lima *et al.*, 2012. Tannins, peptic ulcers and related mechanisms. Int. J. Mol. Sci., 13: 3203-3228.
19. Ma, L. and J. Liu, 2014. The protective activity of *Conyza blinii* saponin against acute gastric ulcer induced by ethanol. J. Ethnopharmacol., 158: 358-363.
20. Tyagi, L., N. Sharma, A. Sharma and S. Ahmad, 2019. Evaluation of synergistic antiulcer activity of *Aegle marmelos* and *Ocimum sanctum* in ulcer induced Wistar rats. Panacea J. Pharm. Pharm. Sci., 8: 1-9.