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THE ANTIULCER EFFECTS OF THE METHANOL EXTRACT OF THE LEAVES OF *ASPILIA AFRICANA* (ASTERACEAE) IN RATS

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Abstract.

Aspilia africana (Asteraceae) is a plant currently used in Cameroon ethnomedicine for the treatment of stomach ailments. The methanol extract of the leaves of *A. africana* was investigated against gastric ulcerations induced by HCl/ethanol and pylorus-ligation. With both methods, the extract inhibited gastric ulcerations in a dose-related manner. Oral administration of the plant extract at the doses of 0.5 and 1 g/kg reduced gastric lesions induced by HCl/ethanol by 79 % and 97 % respectively. The extract at the dose of 1 g/kg reduced gastric lesion in the pylorus ligated rats by 52 % although the gastric acidity remained higher as compared to the control. These findings show that methanol extract of the leaves of *A. africana* possess potent antiulcer properties.

Key words: *Aspilia africana*, antiulcer, rat

Introduction

Aspilia africana (Pers.)C.D. Adams is a perennial herb which belongs to the family of Asteraceae. It is lignous at the base and can grow up to 1.5 m high. The fruits are quadrangular akenes and the leaves are opposite and hairy. The flowers are composed of floret of a yellow glaring and dazzling colour (Adjanohoun *et al*., 1996). The plant is a common secondary formation species in cultivated farms and fallow lands, occurring in occidental Africa (from Senegal to Cameroon) (Iwu, 1993). *A. africana* is widely used in African folk medicine, to stop bleeding, remove corneal opacities, induce delivery and in the treatment of anaemia and various stomach complaints (Iwu, 1993; Adjanohoun *et al*., 1996)

The present study was undertaken to determine the antiulcer potential of the methanol extract of the leaves of *A. africana*, using two experimental gastric ulcer models namely, HCl/ethanol-induction and pylorus ligation.

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Materials and Methods

Animals

The experiments were carried out on Wistar strain male adult rats, aged 12 to 15 weeks and weighing 170 to 200 g. The rats were raised in the animal house of the Faculty of Science, University of Dschang, Cameroon and fed on normal laboratory rat diet (Granules, Navette) with water given *ad libitum.*

Preparation of the plant extract

The plant material was harvested in Dschang, Cameroon in January and identified at the National Herbarium in Yaounde-Cameroon in comparison with the existing Voucher specimen N° 6555/SRF/CAM. The harvested fresh leaves were sun dried and ground into a fine powder. 430 g of the powder were macerated in methanol for 72 hours. The filtrate was concentrated under reduced pressure at 80° C to give 58.5 g of a green water-soluble residue, corresponding to an extraction yield of 13.6 %.

Phytochemical screening

The use of the test of Shinoda help to determine the presence of flavonoids (Markham, 1982), Libermann Buchard's test revealed the existence of sterols and triterpens (Klyne, 1970), while saponins were revealed as described by Hosttetmann et al. (1991).

HCl/ethanol- induced ulcer

Gastric ulceration was induced in 48 hours starved rats using the method of Hara and Okabe (1985). The rats were given the plant extract at the doses of 0.5 and 1 g/kg orally, the positive controls received cimetidine (p.o.), at the dose of 11.5 mg/kg, while negative control received distilled water. One hour 30 minutes after drug treatment, 1 ml of the necrotizing solution (150 mM HCl in 60 % ethanol) was given *per os* to each rat. The rats were killed 1 hour later using an over dose of ether, and the stomach removed and observed for ulcers in the glandular region. The surface area of each lesion was measured and scored as described by Tan et al. (1996). The ulcer index for each rat was taken as the mean ulcer score. The percentage ulcerated surface was calculated as the total area covered by all lesions expressed as a proportion of the total corpus mucosal surface area. The percentage of inhibition (% I) was calculated using the following formula:

$$\% \, I = \frac{(USc - USt).100}{USc}$$

Where USc = ulcer surface area of control and USt = ulcer surface area of test animal.

Pylorus ligated ulcer technique

The rats were given the plant extract (1 g/kg) or cimetidine (11.5 mg/kg) 1 h before pylorus ligation. Six hs after the ligation, the animals were killed and the stomach removed. The gastric contents were collected, centrifuged and the supernatant measured. The ulcer formed in the gastric mucosa were measured and scored as

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described by Shay et al. (1945). The ulcer index, the percentage ulcerated surface and the percentage of inhibition were estimated as described above.

Measurement of gastric acidity

One ml of the total centrifuged gastric contents from each pylorus-ligated rat was analysed for hydrogen ion concentration by titrating against a 0.01N solution of NaOH. The experiment was done in triplicate.

Statistical analysis

Statistical analysis was performed using ANOVA and Duncan's test and significance of difference between treatments was accepted at $p < 0.05$. Data were expressed as mean ± standard error of the mean.

Results and discussion

HCl/ethanol-induced ulcer

Oral administration of the HCl/ethanol solution produced characteristic lesions in the glandular portion of the rat stomach, with a total surface of 180.00 ± 7.18 mm^2. The leaf methanol extract of *A. africana* produced a dose-dependent inhibition of gastric ulceration ranging from 79 % at the dose of 0.5 g/kg to 97 % at the dose of 1 g/kg with a respective ulcer surface area of 38.20 ± 9.07 and 5.25 ± 1.05 mm^2. The mean ulcer index reduced significantly ($p < 0.05$) from 3.51 in negative control to 2.97 and 1.75 for rats receiving 0.5 and 1 g/kg of extract respectively (Table 1).

Table 1. Effects of the leaves methanol extract of *Aspilia africana* on HCl/ethanol-induced gastric lesions in rats.

Treatment	Dose (mg/kg)	Ulcer Index	U S area (mm^2)	% I	% US
Control	-	3.5 ± 0.42	180.0 ± 7.1	0	8.06
Cimetidine	11.5	2.7 ± 0.19[a]	60.0 ± 5.35[a]	66.67	3.34
Extract	500	2.9 ± 0.32[a]	38.2 ± 9.07[a]	78.89	2.02
	1000	1.7 ± 0.54[b]	5.25 ± 1.05[b]	97.22	0.24

Number of rats per group = 7
I = inhibition
US = ulcerated surface
[a]$p< 0.05$; [b]$p< 0.01$ statistically significant relative to control

These results show that the methanolic extract of *A. africana* leaves has protective properties against HCl/ethanol induced ulcers in rats. This method of ulcer induction is being widely used and is a convenient way of assessing anti-ulcer activity of drug (Hara and Okabe, 1985, Tan et al., 1996, 2000). The gastric mucosal protection against irritant substances as HCl/ethanol can be mediated by a number of mechanisms that

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Table 2. Effects of the leaves methanol extract of *Aspilia africana* on pylorus ligation ulceration

Treatment	Dose (mg/kg)	Ulcer index	US area (mm^2)	% I	% US	gastric juice (ml)	Gastric acidity (mEq/l)
Control	-	2.72 ± 0.21	12.00 ± 3.05	0	1.02	8.65 ± 1.41	104 ± 7.01
Cimetidine	11.5	2.29 ± 0.12*	9.00 ± 3.42	25.00	0.66	6.20 ± 0.77*	98.00 ± 12.68
Extract	1000	1.29 ± 0.64*	5.75 ± 2.01*	52.08	0.58	8.13 ± 0.30	113.00 ± 6.41

Number of rats per group = 7
I = inhibition
US = ulcerated surface
*p< 0.05 statistically significant relative to control

include enhanced gastric mucosal defence through increased mucus and/or bicarbonate production, reducing the volume of gastric acid secretion or by simply neutralizing the gastric acidity. It becomes evident that this model of experimental ulcer is unable to indicate the mechanism of action of the extract. In order to have an idea of the possible mechanisms of action of the extract, its antiulcer potency was tested against pylorus ligated – induced ulcer.

Pylorus ligated-induced ulcer

The macromorphological results obtained when ulceration of the gastric mucosa was provoked using pylorus ligation are shown in Table 2. At the dose of 1 g/kg, the methanol extract of the leaves of *A. africana* produced a significant ($p < 0.05$) decrease in ulcer surface area from 12.00 ± 3.05 to 5.75 ± 2.01. The plant extract was unable to affect the volume of gastric juice. The gastric acidity of treated animals (113 mEq/l) was higher than that of the control (104 mEq/l).

This technique revealed that although the methanol leaf extract of *A. africana* significantly reduced gastric ulceration, it was unable to reduce the volume of gastric juice secreted and even tended to increase the gastric acidity. These results suggest that the extract possesses neither anti-secretory potency nor any neutralizing effect. It is known that the cytoprotective action of some antiulcer drugs is mediated by the action of endogenous prostaglandins which promote mucus secretion and play an important role in maintaining mucosal integrity against the actions of various damaging agents (Miller *et al*, 1982; Jain *et al*., 2002). Substances like prostaglandins can thus provide gastric cytoprotection in rats against strong necrotizing irritants without reducing gastric acid secretion. It can be thought that the cytoprotective effect of the extract results from the enhancement of the mucosal barrier through the increase production of prostanglandin. Yamamoto et al. (1992) and Tan et al. (2000) have shown the prostaglandin-like cytoprotective action of some hydroxychalcone and methylene chloride extract of *Bidens pilosa* respectively and the methanol leaf extract of *A. africana* might be producing its antiulcer effects through the same mechanism. The anti-ulcer effects of the methanol extract of the leaves of *Aspilia africana* may be due to the presence of triterpenes in the extract. Some triterpernic compounds such as nimbidin have been shown to possess anti-ulcer activity (Pillai and Santhakumari, 1984). Cimetidine at the dose of 11.5 mg/kg reduced the ulcer lesion induced by HCl/ethanol (67 %) or by pylorus ligation (52 %). Additionally, it was able to reduce

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the volume of gastric juice. This goes to show its effectiveness in preventing gastric ulcer and also confirm its anti-secretory activity. More work is going on in our laboratory on the bio-guided fractionation of the methylene chloride/ methanol extract of the plant.

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