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IJPBS |Volume 3| Issue 1 |JAN-MAR |2013|145-152

Research Article
Biological Sciences

ANTI ULCER ACTIVITY OF *ACACIA FARNESIANA (L.) (AROMA)* *A LESSER KNOWN FOLK - MEDICINAL PLANT*

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ABSTRACT

Aim of the study: *To determine the pharmacological activity relevant to ulcer healing of* ***Acacia farnesiana (aroma)*** *leaf, a folk medicine used in the treatment of Ulcer.*

Materials and Methods: *MEAF extract was done by using soxhelet apparatus and was further subjected to screening of the qualitative analysis of phyto chemicals. Consequently the Methanolic extract of leaf was tested against ulcer induced rats.*

Results: *The methanol extract of leaf (200mg/Kg) has shown greater protection against ulcer damage to the tissue (58.35 compared to 61.12* ***%*** *of RD)*

Conclusion: *After performing of Preliminary Phytochemical screening of* ***Acacia farnesiana (aroma),*** *the Methanol extract of* ***Acacia farnesiana (aroma)*** *was chosen for further analysis, in other hand the chronic study was done in Ethanol induced ulcers of Male Wister albino rats and then the Ulcer Scoring as well as the Biochemical parameters were studied. Consequently we have evaluated the Anti Ulcer activity of MEAF. Hence, it establishes the fact that the Methanol extract of antiulcer activity of* ***Acacia farnesiana (L.) (Aroma)*** *is offering an ineffable support to the folk medicine, which is a traditional medicine for the treatment of ulcers in INDIA.*

KEY WORDS

Anti ulcer, Acacia farnesiana, Folk Medicine, Aroma

1. INTRODUCTION

Peptic ulcer is a conglomerate of heterogenous disorders which manifests itself as a break in the lining of the gastrointestinal mucosa bathed by acid and/or pepsin. NSAIDS ingestion is associated with erosions, petechiae type C gastritis, ulceration, interference with ulcer healing, Ulcer complications and injury to the small and large intestine[16].

Although a number of antiulcer drugs such as H2 receptor antagonists, proton pump inhibitors and cyto protectants are available for ulceration, but all these drugs have side effects and limitations[2]. Herbal medicine considered safer because of the natural ingredients with no side effects[4]. In India, ***Acacia farnesiana (aroma)*** is known as Mulla tumma, Kampu tumma in local area and it is commonly known as Aroma and sweet acacia also. Grown throughout India, and often planted in gardens. If we see its yield, in India and other Eastern countries produce much for local use and Trees begin to flower from the third year, mainly from November to March[5]. The bark of this plant is used as astringent and demulcent. The leaves and roots are used for medicinal purposes. Woody branches used in

India as tooth brushes. The gummy roots also chewed for sore throat. The roots of this plant is also used for the antispasmodic, aphrodisiac, astringent, demulcent, diarrhea, febrifuge, rheumatism, and stimulant[5].The flower infusion of this plant used as a stomachic[9]. It is also used for dyspepsia and neuroses. Mexicans sprinkle powdered dried leaves onto wounds. The flowers are added to ointment, rubbed on the forehead for headache. Green pods are decocted for dysentery and inflammations of the skin and raucous membranes. Colombians bathe in the bark decoction for typhoid. Costa Ricans decoct rhe gum from the trunk for diarrhea, using the pod infusion for diarrhea, leucorrhea, and uterorrhagia. Panamanians and Cubans used the pod to treat conjunctivitis. Cubans use the pod decoction for sore throat. For rheumatic pains, West Indians bind bark strips to the afflicted joint. The root decoction has been suggested as a folk remedy for tuberculosis. The decoction of the root, used in hot baths, is said to help stomach cancer. A plaster, made from the pulp, is said to alleviate tumors[7]. In preliminary phytochemical investigations it has found that the leaves contain lipids, carotenoids, alkaloids, flavonoids and reducing and non-reducing sugars and seven polyphenols (gallic acid, ellagic acid, m-digallic acid, methyl gallate, kaempferol, atomadendrin, and narigenin). Also found narigenin-7-glucoside and naringenin-7-rhamnoglucoside (naringin), as well as naringenin, glucose, and gallic acid [6]. Another Phytochemical compound Quercetin, of this plant is found to be shown antioxidant activity[15]. Some recent reports have indicated that many flavonoids possess antiulcerogenic activity. Oral treatment with the ether fraction of the flavonoid extract demonstrated a good level of gastric protection. Mucous content was increased and accompanied by proportionate increase in proteins and hexosamines[1]. Quercetin, kaempferol, morin, myricetin and rutin when tested were found to inhibit the mucosal content of platelet activating factor (PAF) in a dose dependent manner suggesting that the protective role of these substances may be mediated by endogenous PAF[10]. Flavonoids exhibit several biological effects such as anti inflammatory, anti hepatotoxic and antiulcer actions[3]

In this present study we evaluated the antiulcer activity of MEAF leaf in ulcer induced experimental rats and It is further investigated and confirmed by using bio analytical studies of different antioxidant enzymes by taking the consideration of Phytochemical compounds of ***Acacia farnesiana(aroma).***

2. MATERIALS AND METHODS:

2.1. Plant collection:

The whole plant of ***Acacia farnesiana (Aroma)*** was collected from coastal area, near Chittoor district of A.P. INDIA, during the month of November. It is identified and authenticated.

2.2. Preparation of extract:

The plant of ***Acacia farnesiana (Aroma)*** were shade dried and the leaves reduced to coarse powder in a mortar and pestle. The powdered material obtained was then subjected to successive extraction by hot percolation method using petroleum ether, chloroform and methanol solvents in a soxhelet apparatus. The different extracts obtained were evaporated at 45^0 C to get a semisolid mass. The extracts thus obtained were subjected to phyto-chemical analysis. The percentage yield alcoholic extract was found to be 55.5%w/w and the Methanolic extract was taken for further ***Phytochemical Screening*** Qualitative tests for the presence of plant secondary metabolites such as carbohydrates, alkaloids, tannins, flavonoids, proteins, saponins and glycosides were carried out on extracts using standard procedure studies.

2.3. Animals used:

Male albino Wister rats between 1 to 2 months of age and weighing 125-150g were procured and were maintained as per the guidelines of National Institute of Nutrition (NIN) Animal User's Manual. Animals were acclimatized for 7 days to our animal house, maintained at temperature of 20-24^0 c. The light source in the animal room was regulated with 12 h light period followed by 12 h dark schedule. Two to three animals were housed per cage sized (41× 28 × 14 cm). Paddy husk was used for bedding and on every alternative day bedding was changed and washed thoroughly with water along with Domex, a disinfectant and a detergent. The rats were fed on a standard pellet diet purchased from Sai Durga Feeds and Foods Bangalore, and water ad libitem.

2.4. Experimental Design

The animals were divided into four groups, each consisting of six rats. Group I represented the Normal control group, which received distilled water orally. Groups II represented the Control group, which received Ethanol 1ml/100g.b.w[13]. Groups III received Methanol extract of ***Acacia farnesiana (aroma)*** 200 mg/kg and, Ranitidine, in the dose of 20 mg/kg were administered orally for group IV as reference standard drug. The gastric ulcers were induced in rats by administrating absolute ethanol (95%) (1 ml/100 g b.w.) Orally[13]. After 45 min of Methanol extract and Ranitidine treatment .They were kept in specially constructed cages to prevent coprophagia during and after the experiment. The animals were anaesthetized 1hr latter with anesthetic ether and stomach was incised along the greater curvature and ulceration was scored.

3. ULCER INDEX / SCORING OF ULCER

3.1. MEASUREMENT OF ULCER INDEX:

Stomach mucosa was flushed with saline and lesions in glandular portion were then exposed and examined under a 10x magnifying glass[8]. Ulcer index of each animal was calculated by adding the values and their mean values were determined by the following scoring system [8].

For the macroscopic observations, the number, lengths and severity of ulcers were noted and scored. The ulcer index (U.I.) of each stomach was the sum of its scores. The ulcer index was reported as arithmetic means ± S.E. The significance of differences between means was evaluated by Student's *t* - test for unpaired data. *P*< 0.05, versus control, was taken as significant.

(i) Normal coloured stomach– 0,
(ii) Red colouration - 0.5,
(iii) Spot ulceration – 1,
(iv) Haemorrhagic streak – 1.5,
(v) Ulcers (< 2mm) – 2,
(vi) Ulcers (>2 < 4 mm) perforations – 3.
(v) 3 Ulcers (< 4mm): -4

Percentage inhibition was calculated using the following formula:

$$\% \text{ Inhibition} = \frac{UI_{\text{ulcer control}} - UI_{\text{treated}}}{UI_{\text{ulcer control}}} \times 100$$

4. Sample collection and preparation for biochemical estimations and assays:

4.1. Measurement of gastric secretion and P^H. [11]

The stomach of aspirin induced ulcer rats was carefully excised keeping oesophagus closed and opened along greater curvature and luminal contents were removed. The gastric juice thus collected was centrifuged at 3000 rpm for 10 min and expressed in terms of ml/100 g of body weight. The pH of the supernatant was measured using digital pH meter [11].

4.2. Measurement of Free and total acidity[12]:

Free and total acidity were determined by titrating with 0.01N NaOH using Topfer's reagent

and phenolphthalein respectively as indicators and were expressed as meq/l per 100 g [12].

4.3. Measurement of gastric juice and pH[11]:

The gastric juice was collected was centrifuged at 3000 rpm for 10 min and expressed in terms of ml/100 g of body weight. The pH of the supernatant was measured using digital pH meter.

4.4. Determination of free acidity and total acidity[14]:

The gastric contents were centrifuged at 1000rpm for 10min. 1ml of supernant was diluted with 9ml of distilled water. A volume of 2ml diluted gastric juice was titrated with 0.1N Sodium hydroxide run from a micro burette using 3-4 drops of Topfer's reagent as indicator until canary yellow colour was observed. Volume of NaOH required was noted. This corresponds to free acidity. Further 2-3 drops of phenolphthalein was added and titrated with NaOH until pink colour was restored. This gives total acidity. Free acidity and total acidity is expressed in terms of ml of 0.1N HCl per 100 gms of gastric contents. This is the same as mEq/lit. To obtain this figure multiply the burette reading obtained from titration by 10.

Acidity was calculated by using the formula:

$$\text{Acidity} = \frac{\text{Volume of NaOH X Normality of NaOH X 100}}{0.1} \text{ meq/L/100g}$$

5. RELUTS

ANTI ULCER EFFECT OF MEAF IN ETHANOL INDUCED MODEL

TABLE A ANOVA TEST FOR ANTI ULCER ACTIVITY OF ETHANOL INDUCED MODEL

S.NO	TREATMENT	ULCER INDEX
1.	CONTROL	5.917 ± 0.2386
2.	RANITIDINE	2.167 ± 0.3073
3.	MEAF	2.417 ± 0.3005
F, d f Value		54.569 (2/15)
P Value		P < 0.0001

*** P < 0.01 when compared with Control**

Figure A: Graphical Representation of Ulcer Index

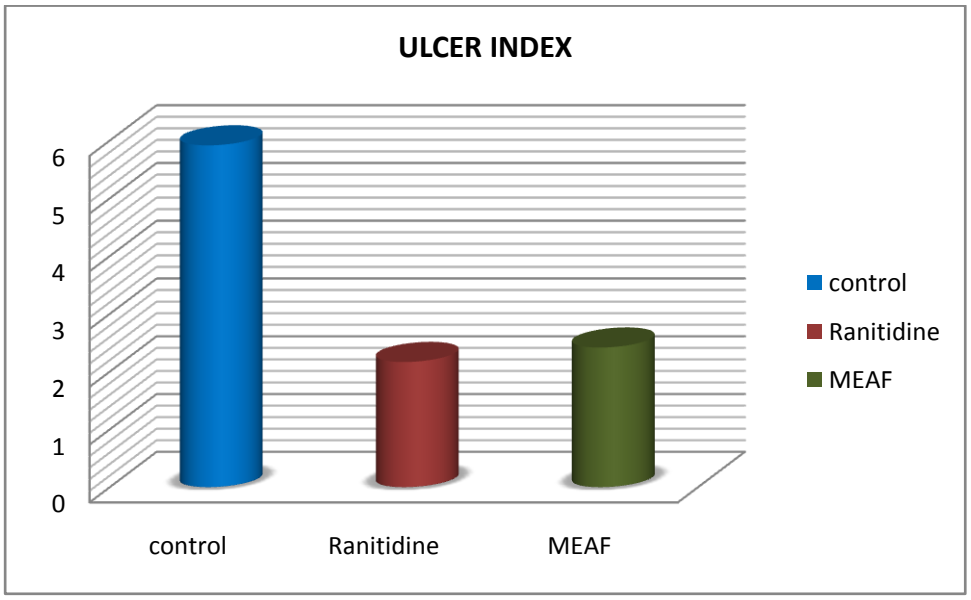

TABLE B ANOVA TEST FOR ANTI ULCER ACTIVITY OF PYLORUS LIGATED ULCER INDUCED MODEL:

S.NO.	Treatment	Volume of Gastric juice(ml)	pH	Free Acidity meq/l/100gm	Total Acidity meq/l/100gm
6.	CONTROL	3.117 ± 0.1579	1.717 ± 0.04773	21.500 ± 0.9916	79.33 ± 1.333
7.	RANITIDINE	1.217 ±0.04773	2.967 ± 0.1229	10.833 ± 0.6009	39.500 ± 1.147
8.	MEAF	1.417 ± 0.06009	2.633 ± 0.1282	12.833 ± 0.6009	49.833 ± 1.078
F, ,d f Value		106.05 (2/15)	37.151 (2/15)	56.547 (2/15)	301.22 (2/15)
P Value		P < 0.0001	P < 0.0001	P < 0.0001	P < 0.0001

*** P < 0.01 when compared with Control**

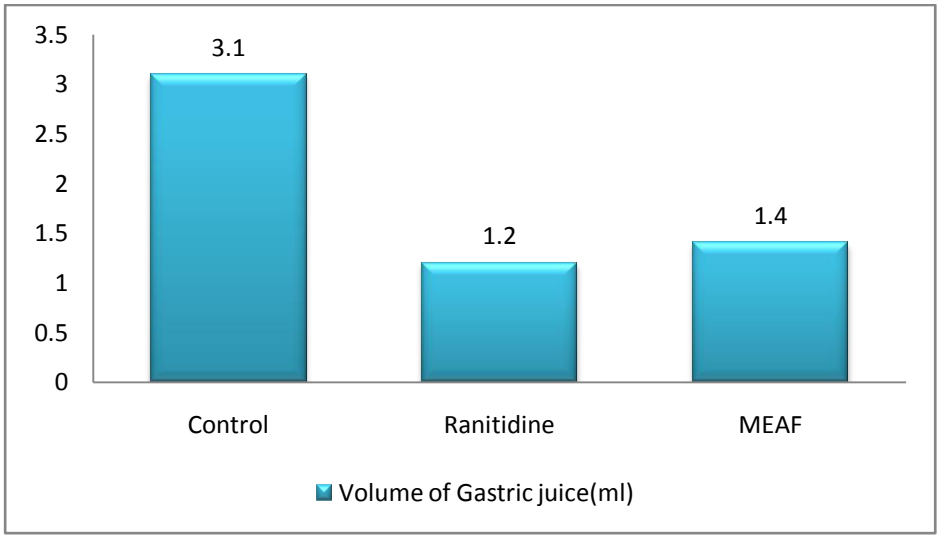

FIGURE B: ANOVA Test for Gastric Juice Volume

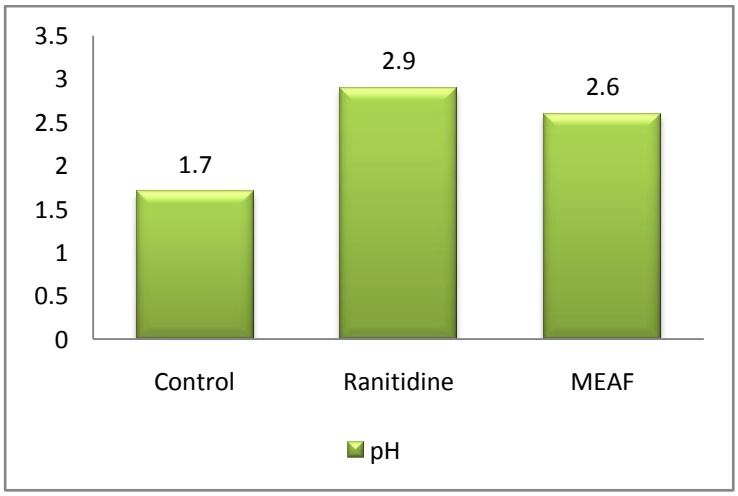

FIGURE C: ANOVA Test for pH

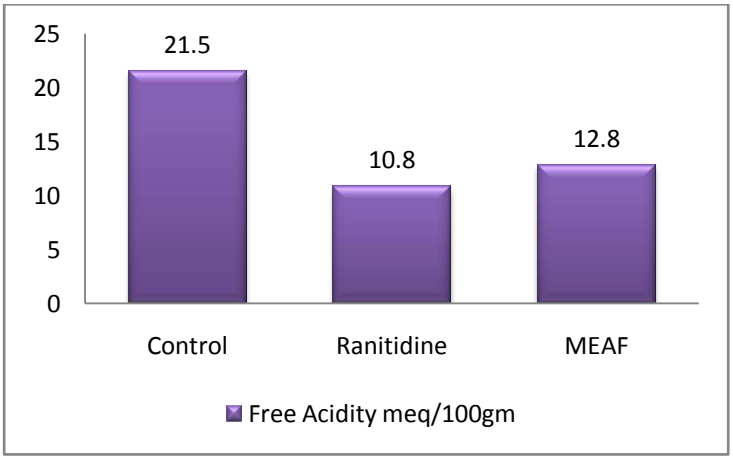

FIGURE D: ANOVA Test for free acidity

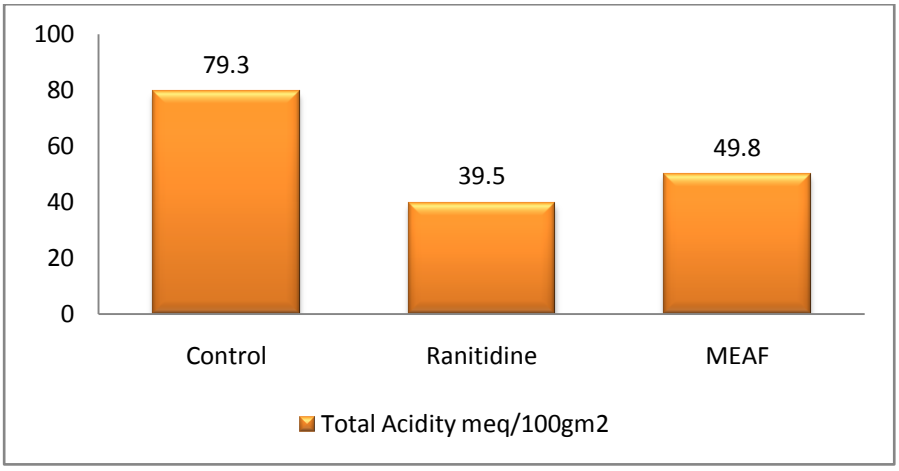

FIGURE E: ANOVA Test for Total acidity

6. DISCUSSION

Oral administration of methanol extract of ***Acacia farnesiana(aroma)*** leaves at a dose of 200 mg/kg exhibited dose dependent inhibition Percentage of 58.35 compared to the ulcer control, proving the anti-ulcer activity of extract whereas ranitidine (20 mg/kg) produced 61.12 % inhibition of ulcer index against Ethanol induced ulcer. MEAF significantly protected gastric mucosa against the damage induced by ethanol and Curative ratio of the MEAF of 200 mg/kg was found to be 90.17%. Oral administration of MEAF 1 hr before the induction of stress reduced the cold restrained stress induced ulcers. The MEAF exhibited a dose dependent inhibition percentage of 58.35 at doses of 200 mg/kg dose. The standard drug Ranitidine showed an inhibition percentage of 61.12.

Ethanol administration (20mg/kg) resulted in the production of gastric mucosal damage. The ulcer index in control animals was (5.917). Methanol extract (2.417) significantly reduced the ulcer index ($p<0.01$) as compared to control. Ranitidine, a standard anti-ulcer drug showed ulcer index (2.167). The results are tabulated in **Table A**.

Pre treatment of rats with ***Acacia farnesiana (aroma)*** extracts produced a dose dependent protection in the ethanol induced ulceration model as compared to control group. However the protection was statistically significant reduced the severity of ulcer and caused a significant reduction of ulcer index in this model. Ranitidine produced significant gastric ulcer protection as compared to control group. Pre treatment with methanol extract of ***Acacia farnesiana(aroma)*** leaves produced significant anti-ulcer effect which can observed by the effect of MEAF on gastric secretion in Ethanol induced ulcer. Gastric juice volume, total and free acidity significantly increased and pH decreased in ulcer control animal in comparison to normal animals. MEAF (200 mg/kg) produced dose dependent effect and decreased gastric juice volume, total and free acidity and increased pH significantly.

Hence it establishes the fact that the Methanol extract of ***Acacia farnesiana(aroma)*** has shown the protection against the ulcers which was induced by the ethanol and it is further recommended to do the in vivo Bio analytical assays, such as enzymatic studies etc.,

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