

ANTIULCER ACTIVITY OF *ACACIA CATECHU* WILLD IN RATSPatankar Rajendra<sup>1</sup>, Devale Titiksh<sup>2</sup>, Pophale Rasika<sup>1</sup>, Gawande Vandana<sup>2</sup>, Raghav Nikhil\*<sup>1</sup><sup>1</sup>Abhinav College of Pharmacy, University of Pune, Narhe, Pune-411041, Maharashtra, India<sup>2</sup>Sinhgad Institute of Pharmacy, University of Pune, Narhe, Pune-411041, Maharashtra, India

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## ABSTRACT

In present work gastric antiulcer activity and in-vitro acid neutralizing capacity of aqueous extract of heartwood of *Acacia catechu* willd (AEAC) was studied. The study was done on female albino rats. Gastric ulcers were induced by pyloric ligation. Ranitidine was used as the standard drug for comparison. Tannins, Catechins and Flavonoids are present in AEAC. The antiulcer property of *Acacia catechu* in pylorus ligation model is evident from its significant reduction in total acidity, number of ulcers. antiulcer property which could be either due to Action on membrane of microorganism, by mucus accumulation, by inhibiting H<sup>+</sup> K<sup>+</sup> (+)-ATPase and by decreasing mucosal hemorrhage and erosion. In conclusion, the present study provide preliminary data on the antiulcer potential of *Acacia catechu* willd heartwood and support the traditional uses of the plant for the treatment of gastric ulcer.

**Keywords:** Antiulcer activity, *Acacia catechu*, heartwood, Pylorus ligation, Ranitidine

## INTRODUCTION

Ulcers are defined histologically as a breach (gap in wall), in the mucosa of the alimentary tract that extends through the muscularis mucosa into the sub mucosa or deeper. Ulcers are to be distinguished from erosions, in which there is epithelial disruption within the mucosa but no breach (gap in wall) on the muscularis mucosa. Peptic ulcers are chronic, most often solitary, lesions that occur in any portion of the gastrointestinal tract exposed to the aggressive action of acid/peptic juices<sup>1-2</sup>. Today, there are two main approaches for treating peptic ulcer. The first deals with reducing the production of gastric acid and the second with re-enforcing gastric mucosal protection<sup>2</sup>.

Although a number of antiulcer drugs such as H<sub>2</sub> receptor antagonists, proton pump inhibitors and cytoprotectants are available for ulceration all these drugs have side effects and limitations Herbal medicine deals with plants and plant extracts in treating diseases. These medicines are considered safer because of the natural ingredients with no side effects<sup>3</sup>.

*Acacia catechu* (Khair, Kaat, and Katthaa) belongs to family Mimosaceae. It has been traditionally used for treatment of diarrhea, dysentery, colitis, piles, ulcers, boils and skin eruptions internally as well as externally<sup>4</sup>. Catechu contains catechuic acid, catechutannic acid (25%-33%), acacatechin (10%-12%), catechu red, quercetin, catechin (2%-12%), epicatechin, phlebotannin (25%-33%), gummy matter, quercitrin, quercetin, fisetin, moisture and ash. Quercitrin is a phenolic flavonoid and catechu of acacia is a psuedotannin. Catechu and epicatechin usually accompany other flavonoids. It also contains the active principles like cyanodanol, tannin and polyphenols<sup>5-6</sup>.

Literature review on *Acacia catechu* shows that it has Antipretic, Antidiarrhoeal, Hypoglycemic, Hepatoprotective, Immunomodulatory, Anti mycotic activities<sup>7-9</sup>. It has also shown potential to inhibit cyclooxygenase and 5-lipoxygenase to reduce inflammation<sup>10</sup>.

In present study we propose the antiulcer activity of *A. catechu* by following possible mechanisms:

- Action on membrane of microorganism,
- By mucus accumulation,
- By inhibiting H<sup>+</sup> K<sup>+</sup>(+)-ATPase and
- By decreasing mucosal hemorrhage and erosion.

**Action on membrane of microorganism**

Catechins mainly (-)-epicatechin which is present in *Acacia catechu* causes little damage to the membrane of the microorganism. The bacterial infections are prolonged due to presence of fibrin-rich

biofilm and the formation of fibrin-rich membranous structure by bacteria will probably be inhibited by "tannins"<sup>11</sup>. Polyphenolics are partially hydrophobic, this may allow them to interact with the bacterial cell wall and lipopolysaccharide interfaces more effectively by decreasing membrane stability<sup>12</sup>.

**By mucus accumulation**

It was supposed that the mucus accumulation is resultant from a possible reaction between tannins and mucopolysaccharides<sup>13</sup>.

**By inhibiting H<sup>+</sup> K<sup>+</sup> (+)-ATPase**

The intensity of inhibitor activity is directly proportional to the number of phenolic hydroxy groups in the Catechins. The inhibition of the enzyme by (-)-epicatechin was competitive with respect to ATP and noncompetitive with respect to K<sup>+</sup>. These findings suggest that the anti-secretory and anti-ulcerogenic effects of Catechins are due to their inhibitory activity on gastric H<sup>+</sup>, K<sup>+</sup> (+)-ATPase<sup>14</sup>.

**By decreasing mucosal hemorrhage and erosion**

In infected animals, *H. pylori* was eradicated in 10 to 36% of the catechin treated animals, with significant decreases in mucosal hemorrhage and erosion<sup>15</sup>. Considering the above possible mechanisms, the present work was undertaken to evaluate antiulcer activity of aqueous extract of *Acacia catechu* in experimental models.

**MATERIALS AND METHODS**

Ranitidine injection (RANTAC INJECTION, 50mg/2ml, RANBAXY) was purchased from local market. All Other chemicals and reagents used were of analytical grade.

**Plant material**

The heartwood of *Acacia catechu* Willd. was collected from the Abhinav Education Society's college of pharmacy campus in Pune, during the month of December 2009. The plant was authenticated by Mr. Raju Singh, Botanist, Botanical survey of India, Pune.

**Experimental animals**

Wistar rats (female) weighing around 140-160g was procured from National institute of Biosciences, Pune. All the animals were place in polypropylene cages in a controlled room temperature and relative humidity in registered animal house (922/ac/05/CPCSEA). The animals were maintained on standard diet and water ad libitum. They were acclimatized to laboratory condition for seven days before commencement of the experiment. Animal experimental studies were conducted according to the guidelines of institutional animal ethical committee and approval was obtained for the protocol designed.

**Preparation of plant extract**

The dried heartwood of *Acacia catechu* weighing about 100g was taken and about 400ml distilled water was added to it into a 1000ml beaker. Then it was kept for boiling to around 15 minutes and it was then kept for maceration for 24 hours. After 24 hours it was then filtered through a suction pump to get the concentrated filtrate which was evaporated to get a semisolid extract and was stored in fridge. Then the residue remained from first maceration was again kept for maceration for 24 hours, filtered, evaporated as above mentioned to get an concentrated extract. These extract was also kept in fridge. The dried extract was used for further study. The yield of the extract was found to be 14.3%.

**Preliminary Phytochemical screening**

Various Phytochemical tests were carried out on the *Acacia catechu* heartwood extract to detect the presence of Tannins, Catechins, Flavonoids and other compounds<sup>4, 16</sup>.

**ASSESSMENT OF ANTI-ULCER ACTIVITY****Pyloric ligation induced ulcer**

Albino rats (Female) were divided into four groups of six animals each. Animals were fasted for 24 h before the study, but had free access to water. Animals in the control group received only 3% gum acacia solution (5ml/kg). Aqueous extract of *A. catechu* at 200 mg/kg, p. o. were given to the animals in the treatment group. Ranitidine (100mg/kg) was used as standard. After 1h of drugs treatment, they were anaesthetized with the help of anesthetic ether; the abdomen was opened by a small midline incision below the xiphoid process. Pyloric portion of the stomach was slightly lifted out and ligated according to method of Shay et al., avoiding traction to the pylorus or damage to its blood supply. The stomach was replaced carefully and the abdominal wall was closed by interrupted sutures. Rats were sacrificed by an over dose of anesthetic ether after four hours of pyloric ligation. The abdomen was opened, cardiac end of the stomach was dissected out and the contents were drained into a glass tube. The volume of the gastric juice was measured and centrifuged at 2000 rpm for 10 min. From the supernatant, aliquots (1 ml of each) were taken for the determination of pH, total and free acidity. The inner surface of free stomach was examined for gastric lesions<sup>17-18</sup>.

**Determination of pH**

An aliquot of 1ml gastric juice was diluted with 1ml of distilled water and pH of the solution was measured using pH meter<sup>18</sup>.

**Determination of total acidity**

An aliquot of 1ml gastric juice diluted with 1ml of distilled water was taken into a 50ml conical flask and two drops of phenolphthalein indicator was added to it and titrated with 0.01N NaOH until a permanent pink color was observed. The volume of 0.01N NaOH consumed was noted. The total acidity is expressed as Meq/l by the following formula

$$\text{Total Acidity} = n \times 0.01 \times 36.45 \times 1000$$

Where n is volume of NaOH consumed, 36.45 is molecular weight of NaOH, 0.01 is normality of NaOH, 1000 is the factor (to be represented in liter)<sup>18</sup>.

**Macroscopic evaluation of stomach**

The stomach of the animals were opened along the greater curvature, rinsed with saline to remove gastric contents and blood clots and examined by a 5X magnifier lens to assess the formation of ulcers. The number of ulcers was counted<sup>18</sup>.

**Statistical Analysis**

The results were expressed as the mean  $\pm$  SEM for each group. Statistical differences were evaluated using a One-way analysis of variance (ANOVA) followed by Dunnet's t-test. Results were considered to be statistically significant at  $P < 0.05$ .

**RESULTS****Phytochemical screening**

The outcome of the preliminary phytochemical screening of the extract showed the presence of Flavonoids, tannins and Catechins

**Effect of AEAC on Pyloric ligation induced gastric ulcers**

Effect of Aqueous extract of *A. catechu* on pyloric ligation induced ulceration is shown in Table 1. It indicates significant decrease in Volume of gastric juice, pH, Total acidity and Number of ulcers.

**DISCUSSION**

Although in most of the cases the etiology of the ulcers is unknown, it is generally accepted that they are a result of an imbalance between aggressive factors and the maintenance of mucosal integrity through endogenous defensive mechanisms<sup>19</sup>.

To regain the balance, different therapeutic agents including plant extracts may be used<sup>20-21</sup>. *A. catechu* extract is one such herbal drug used in the present study primarily to evaluate the anti-ulcerogenic or ulcer preventive potency in pylorus ligation induced ulcers in rats.

Pylorus ligation induced ulcers are due to auto digestion of the gastric mucosa and breakdown of the gastric mucosal barrier<sup>22</sup>. These factors are associated with the development of upper gastrointestinal damage including lesions, ulcers and life threatening perforation and hemorrhage. Prostaglandin E2 and I2 are predominantly synthesized by the gastric mucosa and are known to inhibit the secretion of gastric acid and stimulate the secretion of mucus and bicarbonate. Hydrophobic surfactant - like phospholipids secretion in the gastric epithelial cells is also stimulated by the prostaglandin<sup>23</sup>. In addition, Brodie<sup>24-25</sup> also showed development of gastric ulcers in pyloric ligation model. Volume of gastric secretion is an important factor in the production of ulcer due to exposure of unprotected lumen of the stomach to the accumulating acid<sup>21</sup>.

The antiulcer property of *A. catechu* in pylorus ligation model is evident from its significant reduction in total acidity, number of ulcers. *A. catechu* treated animals significantly inhibited the formation of ulcers in the pylorus ligated rats and also decreased both the concentration and increased the pH, it is suggested that *A. catechu* can suppress gastric damage induced by aggressive factors.

**CONCLUSION**

The extracted product gave significant tests and it was confirmed that Tannins, Catechins and Flavonoids are present in *Acacia catechu* heartwood extract. The results of the present study suggest that the aqueous extract of *Acacia catechu* heartwood may be beneficial in the treatment of gastric lesions. Further studies to identify the active moieties and elucidation of the mechanism of action are recommended.

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TABLE 1: EFFECT OF ACACIA CATECHU HEARTWOOD EXTRACT ON GASTRIC ULCERS

Groups	Dose ml/kg	Volume of gastric juice (ml/4hrs)	pH	Total acidity (meq/l)	Number of ulcers
Control (Gum acacia)	5	3.550 ± 0.1310	3.83 ± 0.085	1276 ± 34.58	1.778 ± 0.3643
Test	200	1.825 ± 0.0458*	4.39 ± 0.13*	918.5 ± 31.36	0.888 ± 0.2003* (AEAC)
Standard (Ranitidine)	100	1.433 ± 0.0918*	5.08 ± 0.088*	750.9 ± 39.26	0.666 ± 0.1667*
			p	<0.05	

\* p<0.05 when compared to the control group.

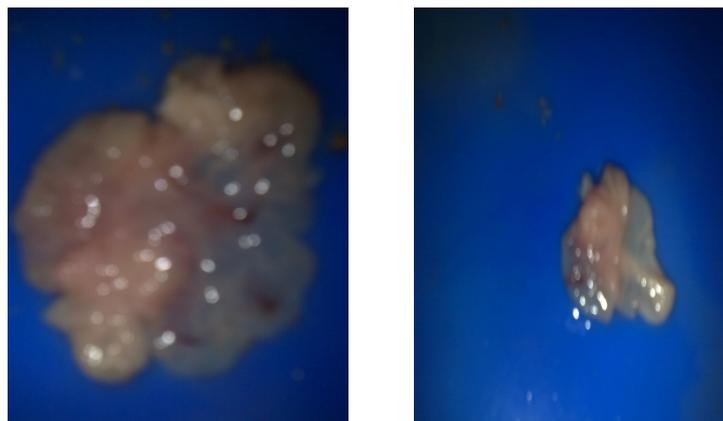


Figure 1(a) Ulcers on stomach of control animal. (b) Ulcers on stomach of test animal.

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