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Evaluation of anti-ulcer activity of ethanolic extract of *Abutilon Indicum* (Linn.) sweet leaves

K Ponnudurai[1*], K Prabhu[2], D Jebasingh[3] and D Prabu[3]

[1]*Department of Pharmacology, Nandini Nagar Mahavidyalaya College of Pharmacy, Nawabganj, Gonda, Uttar Pradesh, India*
[2]*Department of Pharmacognosy, Nandini Nagar Mahavidyalaya College of Pharmacy, Nawabganj, Gonda, Uttar Pradesh, India*
[3]*Department of Pharmacology, C. L. Baid Metha College of Pharmacy, Thorapakkam, Chennai, Tamilnadu, India*

ABSTRACT

The active constituent of leaf extract of the plant (Abutilon indicum) contains various components such as essential oils in particular eugenol which is reported to possess antioxidant potential. Eugenol is the one of the constituents of plant under investigation. Therefore based on the above facts, it is clear that no scientific studies have been carried out in Abutilon indicum regarding the anti-inflammatory and gastro protective activity, the present study has been carried out to investigate and evaluate the anti-ulcer activity of ethanolic extract of Abutilon indicum (Linn.) Sweet leaves.

Key words: Leaves, *Abutilon indicum*, Eugenol, ethanolic extract, gastro protective activity.

INTRODUCTION

A review of literature shows that the following works have been carried out on the plant *Abutilon indicum* (Linn.) Sweet. Especially several pharmacological and phytochemical works have been carried out on the leaves, roots and seeds of *Abutilon indicum* (Linn.) sweet. In particular, phytochemical, pharmacological and pharmacognostical works have been exhaustively done on the leaves of *Abutilon indicum* (Linn.) Sweet. The list of works carried out on the various parts of the plant are as follows.The seeds contain water soluble mucilage and crude protein. It also yields a pale-yellow, semi-drying oil which consists of palmitic acid, stearic acid, oleic acid, linoleic acid and linolenic acid [1, 2]. The petals contain cyanidin-3-rutinoside, gossypetin-8-glucoside and gossypetin-7-glucoside [3]. Eudesmol, geraniol and caryophyllene identified in essential oil of flowers.Amino acids, glucose, fructose, vitamin-C and galactose, β -pinene, caryophyllene, caryophyllene oxide, cineole, geraniol, geranyl acetate, elemene, eudesmol, farnesol and borneol identified in oil [4]. Flavonoids have also been isolated [5]. Fixed oil and gallic acid were isolated from roots [6]. Alkaloids, flavonoids, sterols, saponins, tannins,

triterpeniods and glycosides have been identified from different types of extracts of leaves [7].A water soluble galactomannan has been isolated from the seeds containing D-galactose and D-mannose in 2:3 molar ratio. The seed-gum has branched structure consisting of linear chain β-D (1→4) linked mannopyranosyl units, some of which are substituted at 0-6 by two α-D (1→6) galactopyranosyl units mutually linked glycosidically as end groups. An ethanolic extract of the plant showed anticancer and hypothermic activity, and affected the central nervous system in mice [8]. Alcoholic and water extracts of *Abutilon indicum* leaves showed significant hypoglycemic effect in normal rats from the 4^{th} hour to 8^{th} hour by approximately 23%. Antidiarrhoeal activity of leaf extracts of *Abutilon indicum* using petroleum ether, methanol and distilled water was evaluated by gastro intestinal motility, castoroil-induced diarrhoea and prostaglandin E_2 – induced enteropooling in rats [9].The aqueous extract of leaves of *Abutilon indicum* was tested for hepatoprotective activity against carbon tetrachloride and paracetamol-induced hepatotoxicities in rats [10]. Preliminary phytochemical tests demonstrated the presence of steroids in petroleum ether and benzene extracts which were found to induce dose dependent CNS depression. Similarly, these extracts showed very good analgesic property, whereas alcoholic and aqueous extracts failed to show analgesic activity but all the extracts of the leaves were found to possess hypoglycemic activity [11]. The aqueous extract of the leaves of *Abutilon indicum* showed significant hepato-protective activity against carbon tetrachloride treated rats [12].

Traditional uses

Leaves are cooked and eaten for bleeding piles. Their extract is used as a diuretic, demulcent and as an emollient fomentation; along with ghee as a remedy for diarrhoea. Decoction of leaves used as mouthwash in toothache and tender gums, also useful in gonorrhoea, inflammation of bladder and for enema and vaginal infection. The leaves are also applied on wounds and ulcers [13]. Bark is used as astringent and diuretic [14]. Flowers are eaten raw. They are reported to be employed as an application to boils and ulcers. The powdered flowers are eaten with ghee as a remedy in blood vomiting and in cough .Seeds are considered laxative in piles, used in treating cough and are also distinctly useful in gonorrhoea, gleet and chronic cystitis .Root is used as demulcent and diuretic, prescribed in fever, chest infection and urethritis [15].

MATERIALS AND METHODS

Fine chemicals used in these experiments were obtained from Sigma Chemical Company, U.S.A. and all other analytical grade chemicals were obtained from S.D. Fine Chemical Ltd., Mumbai.

Plant materials

The leaves of *Abutilon indicum* (Linn.) Sweet were collected from the region of Chennai, Tamilnadu, in the months of March/April. The plant was identified and authenticated by Research Officer (Pharmacognosy), Central Research Institute (Siddha), Arumbakkam, Chennai-106. A voucher specimen of the plant was deposited at the Department of Pharmacognosy for further reference.

Extraction and preparation of test sample

The freshly collected leaves were cut into small pieces and 100 gm was soaked in ethanol (90% v/v) for 7 days at room temperature with occasional shaking. The container was kept closed throughout the maceration process. At the end of maceration the viscous extract, after filtration was kept in the refrigerator. The extract was then subjected to preliminary qualitative tests [16, 17] to identify the phyto constituents present in the leaf extract. The ethanolic extract was administered to the animals by suspending each time in 1% SCMC.

Experimental animals

Adult wistar rats either sex weighing 180-250 gms were used in the pharmacological and toxicological studies. The inbred animals were taken from the animal house in C.L.Baid Metha College of Pharmacy, Thoraipakkam, Chennai-96. The animals were maintained in well-ventilated room, temperature was maintained at 22 ± 1^{o}C with humidity at 55 ± 5 %. They were fed balanced rodent pelleted diet from Poultry Research Station, Nandanam, Chennai-35, and tap water *ad libitum* throughout the experimental period. The animals were housed for one week, prior to the experiments to acclimatize to laboratory temperature. The experimental protocol was approved by the Institutional Animal Ethics Committee of C.L.Baid Metha College of Pharmacy, Chennai, Tamilnadu, IAEC Ref No: IAEC XIV-20/CLBMCP/2005-2006.

Acute oral toxicity study

The procedure was followed by using OECD 423 guidelines (Organization of Economic Cooperation and Development) (Acute Toxic class method) [18]. The acute toxic class method is a stepwise procedure with 3 animals of a single sex per step. Depending on the mortality and/or moribund status of the animals, on the average 2-4 steps may be necessary to allow judgment on the acute toxicity of the test substance. This procedure results in the use of a minimal number of animals while allowing for acceptable data-band scientific conclusion.

The method used defined doses (5, 50, 300, 2000 mg/kg body weight) and the results allow a substance to be ranked and classified according to the Globally Harmonized System (GHS) for the classification of chemical, which cause acute toxicity.

Six female Wistar rats weighing between 180–250 gm were used for study. The starting dose level of ethanolic extract of *Abutilon indicum* (Linn.) Sweet leaf was 2000 mg/kg body weight p.o as most of the crude extracts possess LD_{50} value more than 2000 mg/kg in b.w.p.o. So 2000mg/kg was used as starting dose. Dose was administered to the rats, which were fasted over night with water *ad libitum* and food were withheld for a further 3-4 hours after administration (p.o) of drugs and observed for signs of toxicity. The same dose was once again tried with another three rats and observed for signs of toxicity.

Body weight of the rats before and after treatment were noted and any changes in skin and fur, eyes and mucous membranes and also respiratory, circulatory, autonomic, central nervous systems, somatomotor activity and behavior pattern were observed and also signs of tremors, convulsions, salivation, diarrhoea, lethargy sleep and coma were noted. The onset of toxicity and signs of toxicity were also noted.

Aspirin plus pylorus ligation model [19]

Wister rats of either sex weighing 180 to 250 gms were divided into five groups of six animals each. Animals were placed in cages with grating floor to avoid coprophagy in fasting period.

Group I - *Received 1% SCMC 10 ml/kg b.w. (p.o)*
Group II - *Received Aspirin 200 mg/kg b.w. (p.o) suspended in 1% SCMC*
Group III - *Received EEAI 100 mg/kg b.w. (p.o) suspended in 1% SCMC*
Group IV - *Received EEAI 200 mg/kg b.w. (p.o) suspended in 1% SCMC*
Group V - *Received Ranitidine 50 mg/kg b.w. (p.o) suspended in 1% SCMC*

Aspirin (200 mg/kg) was administered in non fasted rats once daily for five days. Ranitidine (50 mg/kg) and EEAI (100, 200 mg/kg) were administered orally to the respective treatment groups 30 min before each Aspirin treatment whereas the control group received only 1% SCMC, and was fasted for 36 hours, On the sixth day, immediately after Aspirin treatment pylorus ligation

was done under ether anesthesia. Four hours after pylorus ligation, the animals were sacrificed by giving overdose of ether. The stomach was dissected out after tying the esophageal end. The stomach was cut open along the greater curvature and the contents drained into a small beaker, centrifuged then subjected to analysis for following acid secretary and biochemical parameter. The mucosa was flushed with saline and the stomach was pinned on a frog board and the ulcer score was calculated.

Estimation of Biochemical Parameter

Determination of gastric volume [20]

After sacrificing the rat, the stomach portion was removed, the gastric contents are transferred into a centrifuge tube, and centrifuged at 1000 rpm for 10 minutes, the supernatant liquid was then transferred to a measuring cylinder, and volume was measured.

Determination of pH of the gastric content [21]

1 ml of the gastric juice was collected, and pH was directly measured by using a digital pH meter.

Determination of free acidity and total acidity [22]

1 ml of gastric juice was pipetted into a 100 ml conical flask. 2 to 3 drops of Topfer's reagent was added and titrated with 0.01 N sodium hydroxide (which was previously standardized with 0.01 N of oxalic acid) until all traces of the red colour disappears and the colour of solution was yellowish orange. The volume of alkali added was noted. This volume corresponds to free acidity, then 2 or 3 drops of phenolphthalein solution was added and titration was continued until a definite red tinge reappears. Again the total volume of alkali added was noted. This volume corresponds to total acidity. Acidity was calculated by using the formula:

$$\text{Acidity} = \frac{\text{Vol.of NaOH} \times \text{Actual normality of NaOH} \times 100}{0.1} \text{ mEq/l/100g}$$

Estimation of total proteins [23]

The dissolved protein in gastric juice was estimated in the alcoholic precipitate obtained by adding 90% of alcohol with gastric juice in 9:1 ratio respectively. Then 0.1ml of alcoholic precipitate of gastric juice was dissolved in 1 ml of 0.1 N NaOH and from this 0.05ml was taken in another test tube. To this 4 ml of alkaline mixture was added and kept for 10minutes. Then 0.4 ml of phenol reagent was added and again 10 minutes was allowed for colour development. Reading was taken against blank prepared with distilled water at 610 nm. The protein content was calculated from standard curve prepared with bovine albumin and has been expressed in terms of µg/ml of gastric juice.

Estimation of total hexoses [24]

To 0.4 ml of hydrolysate, 3.4 ml of orcinal reagent was added. The mixture was then heated in the boiling water bath for 15 minutes. This was then cooled under running tap water and intensity of the colour was read at 540 nm against blank by using distilled water instead of hydrolysate. Total hexoses content was determined from the standard curve of D (+) galactose – mannose and has been expressed in µg /ml of gastric juice.

Estimation of Hexosamine [25]

0.5 ml of hydrolysate fraction was taken. To this 0.5 ml of acetyl acetone reagent was added. The mixture was heated in boiling water bath for 20 minutes then cooled under running tap

water. 1.5 ml of 90% alcohol was then added followed by an addition of 0.5 ml of Ehrlich's reagent. The reaction was allowed to take place for 30 minutes. The colour intensity was measured at 530 nm against blank prepared by using distilled water instead of hydrolysate. Hexosamine content of the sample was determined from the standard curve prepared by using D (+) glucosamine hydrolchloride and concentration has been expressed in µg /ml of gastric juice.

Estimation of fucose [26]
The method was carried out by using three test tubes. In one test tube 0.4 ml of distilled water was taken to serve as control and in each of the other two test tubes 0.4 ml of hydrolysates was taken. To all three test tubes 1.8 ml of sulphuric acid: water (6:1) was added by keeping the tubes in the ice-cold water bath to prevent breakage due to strong exothermic reaction. This mixture was then heated in boiling water bath for exactly 3 minutes. The tubes were taken out and cooled. To the blank and to one of the hydrolysate containing tube (unknown) 0.1 ml of cysteine reagent was added while cysteine reagent was not added to the last tube containing the hydrolysate (unknown blank). It was then allowed to react for 40 minutes for completion of the reaction. The reading was then measured at 396 and 430 nm. This was read against standard curve prepared with D (+) fucose. The fucose content is expressed in terms of µg /ml of gastric juice.

Estimation of Sialic Acid [27, 28]
To 0.5 ml of the hydrolysate in 0.1 N sulphuric acid, 0.2 ml of sodium periodate was added and mixed thoroughly by shaking. A time of 20minutes was allowed to elapse before the addition of 1ml of sodium arsenite solution to this mixture. The brown colour produced was disappeared by shaking. Then 3 ml of thiobarbituric acid was added, 4.5 ml of cyclohexanone was added and thorough shaking was done for 15 seconds, till all the colour was taken up by the cyclohexanone supernatant. The mixture was centrifuged to get a clear pink layer of cyclohexanone. The supernatant was pipetted out and intensity of colour was measured at 550 nm. The sialic acid content of the sample was determined from the standard curve of sialic acid and has been expressed in µg /ml of gastric juice.

Estimation of Total carbohydrates [29]
The dissolved mucosubstance in gastric juice were estimated in alcoholic precipitate obtained by adding 1ml of gastric juice to 9 ml of 90% alcohol and the mixture was kept for 10 minutes and the supernatant was discarded. The precipitate separated was dissolved in 0.5 ml of 0.1 N sodium hydroxide. To this 1.8 ml of 6 N HCl was added. This mixture was hydrolysed in the boiling water bath for 2 hours. The hydrolysate was neutralized by 5 N sodium hydroxide using phenolphthalein as indicator and used for the estimation of total hexoses and hexosamine.

The statistical analysis of various studies were carried out using student 't' test and analysis of variance (ANOVA) followed by Dunnett's 't' test, $p < 0.05$ were considered as significant.

RESULTS AND DISCUSSION

Acute oral toxicity study
The acute oral toxicity study was done according to the OECD guidelines 423 (Acute toxic class method). A starting dose of 2000 mg/kg body weight/p.o of EEAI was administered to 3 female rats, observed for three days. There was no considerable change in body weight before and after treatment of the experiment and no signs of toxicity were observed. When the experiments were repeated again with the same dose level, 2000 mg/kg p.o of ethanolic extract of *Abutilon indicum* for 3 days more, and observed for 14 days, no changes were observed from the first set of

experiment. LD_{50} cut off mg/kg body weight was observed as X (unclassified) and Globally Harmonized System (GHS) and comes under X (Unclassified).

Aspirin plus pylorus ligation model
Acid secretary parameters
pH
The pH level was significantly increased (P<0.01) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in pH level, when compared to group-II animals.

Ulcer Score
The ulcer score was significantly increased (P<0.001) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in ulcer score, when compared to group-II.

Gastric Volume
The gastric volume was significantly increased (P<0.01) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in gastric volume level, when compared to group-II.

Free Acidity
The free acidity was significantly increased (P<0.01) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in free acidity, when compared to group-II.

Total acidity
The total acidity was significantly increased (P<0.01) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in total acidity, when compared to group-II.

Total acid output
The total acid output was significantly increased (P<0.01) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in total acid output level, when compared to group-II animals (Table 1).

Total protein
The total protein was significantly increased (P<0.05) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) decrease in total protein level, when compared to group - II animals.

Total hexose
The total hexose was significantly decreased (P<0.001) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.001) increase in total hexose, when compared to group-II.

Hexosamine
The hexosamine was significantly decreased (P<0.05) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.05) increase in hexosamine, when compared to group-II.

Fucose
The fucose was significantly decreased (P<0.05) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.05) increase in fucose, when compared to group-II.

Sialic acid
The sialic acid was significantly decreased (P<0.001) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine level showed a significant (P<0.001) increase in sialic acid, when compared to group-II.

Total carbohydrate
The total carbohydrate was significantly decreased (P<0.05) in the group-II animals, when compared to control group-I. Administration of EEAI and Ranitidine showed a significant (P<0.05) increase in total carbohydrate level, when compared to group-II (Table 2).

Aspirin is a chemical which induces the mucosal damage in the stomach (Figure 1). It reduces the enzymatic levels of total carbohydrate (hexose, hexosamine, fucose and sialic acid) and increases the level of gastric volume, pH, ulcer score, total acidity and total protein. Aspirin plus pylorus ligated group showed significant increase in the ulcer score and acid secretary parameters like the volume of gastric content, total acidity and total acid output [30].

Pylorus ligation model proposed that the digestive effects of accumulated gastric juice and interference in gastric blood circulation are responsible for the induction of ulcers [31, 32].

Increased gastric secretion is also implicated in the causation of gastric ulcer by anti-inflammatory agents [33].

Mucin is a viscous glycoprotein, an important pre-epithelial factor that acts as the first line of defence against ulcerogens. Increase in mucin was due to significant increase in individual muco polysaccharide like sialic acid, hexose, hexosamine and fucose leading to significant increase in total carbohydrates. The mechanism by which the extract augments the mucous secretion is uncertain. The increase in protein content of the gastric juice in untreated ulcer group indicates the damage to the gastric mucosa as a result of which plasma protein leaks into gastric juice. EEAI showed significant reduction in protein concentration, which indicates strengthening of the gastric mucosa, therefore prevents the entrance of plasma protein into gastric juice [34].

Increase in TC:P ratio of the gastric juice indicates the protection of the ulcer where as decrease in the ratio indicates ulceration of stomach. Increased mucin secretion by the gastric mucosal cells can prevent the gastric ulceration by several mechanisms including lessening stomach wall friction during peristalsis as well as acting as effective barrier to the back diffusion of hydrogen ions [35].

The centrally-induced vascular disturbance of mucosal capillaries is being implicated in restraint-induced gastric bleeding. The significant decrease in score of intensity of intraluminal bleeding caused by EEAI in the present study can be partly attributed to their antioxidant potential. The specific pathophysiologic mechanism involved in stress-induced ulcers could be ultimate multifactorial impairment of mucosal defence system. An increase in gastric acid secretion, reduction of gastric mucus and alteration in the microvasculature of the gastric mucosa play a major role in the pathogenesis of stress-induced ulcers.

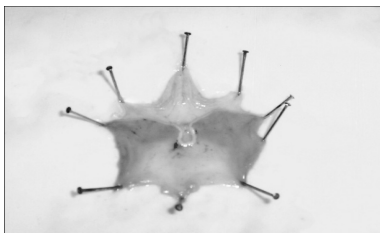
Reactive oxygen species especially $^{\bullet}OH$ plays a major role in oxidative damage of gastric mucosa in almost all forms of gastric ulcer. Increased lipid peroxidation and increased protein carbonyl content are the characteristic features of $^{\bullet}OH$–mediated oxidative damage of the gastric mucosa during ulceration. EEAI is effective in blocking oxidative damage by preventing lipid peroxidation. This is further substantiated by its unique role in scavenging the increased level of endogenous $^{\bullet}OH$ thereby limiting the oxidative stress and associated damage. It also protects the mucosa by preventing adhered mucous depletion [36].

Stress plays an important role in etiopathology of gastro-duodenal ulceration. Increase in gastric motility, vagal over activity; mast cell degranulation; and decreased prostaglandin synthesis are involved in genesis of stress induced ulcers. Complex neurochemical mechanisms are involved in the organism's biological response to noxious stimuli like stress. The pathologic alterations occur with the changes in the synthesis, actions and degradation of hormones, neurotransmitters and neuro modulators.

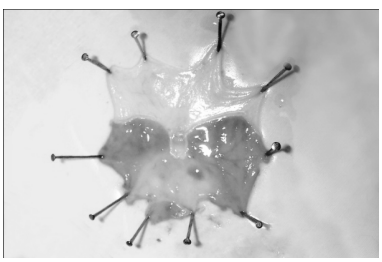
Free radicals affect lipids by initiating peroxidation. Superoxide (O^{-}_{2}), hydrogen peroxide (H_2O_2) and hydroxyl radical ($OH^{\bullet}$) are important ROS causing tissue damage, and lipid peroxide level is an indicator for the generation of ROS in the tissue. The higher lipid peroxidation catalase level and SOD levels indicated increased production of O^{-}_{2} within the tissue, as increased O^{-}_{2} level was thought to increase the concentration of cellular radical level. These radicals functioned in concert to induce cell degeneration via peroxidation of membrane lipids, breaking of DNA strands and denaturing cellular proteins. This effect was significantly reversed by prior administration of EEAI providing a close relationship between free radical scavenging activity [37, 38].

Numerous experimental studies show that exposure of gastric mucosa to potentially noxious environment results in little or no damage, as long as adequate blood flow is maintained. Blood flow contributes to protection by supplying the mucosa with oxygen, bicarbonate, and nutritious substances, and by removing carbon dioxide, hydrogen ions, and toxic agents diffusing from the gastric lumen. Gastric hypoxia, resulting in accumulation of H^{+} within the gastric mucosa, leads to mucosal acidification and development of gastric ulcers.

Figure 1. Gross pathology of the rat stomach after the exposure to Aspirin plus pylorus ligation



Group I



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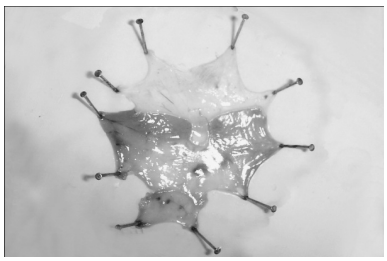

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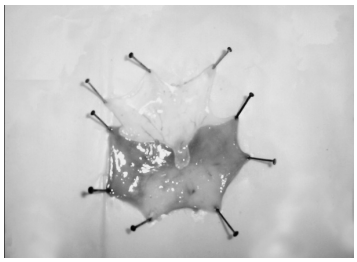

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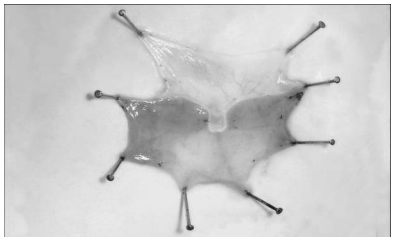

Group V

Table 1. Effect of *Abutilon indicum* leaf extract on ulcer core, pH, gastric volume, free acidity, total acidity and total acid out put

Groups	Ulcer score	pH	Gastric volume (ml/100g)	Free acidity (mEq/l/100g)	Total acidity (mEq/l/100g)	Total acid out put (mEq/100g)
Normal control	3.11 ± 0.428	4.13 ± 0.147	3.283 ± 2.13	58.83 ± 0.70	68.5 ± 1.02	223.75 ± 3.46
Positive control	25.5 ± 0.43*	1.62 ± 0.01*	4.28 ± 0.10*	68.33± 0.76*	78.05± 1.54*	333.85 ±6.53*
EEAI 100 mg/kg	12.33 ± 0.49*	3.33 ± 0.08*	3.083 ± 0.06*	51.16± 0.79*	65.83± 2.16*	202.9± 3.45*
EEAI 200 mg/kg	6.50 ± 0.44*	3.90 ± 0.05*	2.56 ± 0.07*	47± 0.894*	60.16± 2.58**	154.28 ± 3.54*
Ranitidine (50 mg/kg)	5.33 ± 0.45**	4.017 ± 0.05**	2.11 ± 0.07*	42.16 ±0.70**	57.14±3.16**	120.55±3.72**

*Values represented as mean ± SEM of 6 animals, * – p< 0.01, ** – p< 0.001, EEAI –Ethanolic leaf extract of Abutilon indicum*

Table 2. . Effect of *Abutilon indicum* leaf extract on total protein, hexose, hexosamine, fucose, sialic acid, total protein and total carbohydrate-protein ratio

Groups	Total protein (µg/ml)	Hexose (µg/ml)	Hexosamine (µg/ml)	Fucose (µg/ml)	Sialic acid (µg/ml)	Total carbohydrates (µg/ml)	TC : P ratio
Normal control	255.55±11.11	435.5±23.43	239.56±3.21	70.77±1.23	69.63 ± 1.60	815.46±4.04	4.01 ± 0.121
Positive control	455.55±14.05*	241.5±8.99*	151.5±2.29*	103.44±0.66*	26.25± 0.64*	522.69±2.30*	1.14 ± 1.098*
EEAI 100 mg/kg	349.99±14.27*	354.16±11.93*	178.83±1.62*	90.73±1.06*	43.74± 0.76*	667.49± 5.19*	1.31 ± 0.016*
EEAI 200 mg/kg	394.44±15.90*	380.22±6.60**	190.43±1.71*	82.61±1.24*	49.66±0.61**	702.27 ± 2.88**	1.78 ± 0.138*
Ranitidine (50 mg/kg)	311.10±13.05**	394.66±7.74**	199.61±2.25*	79.40±1.01*	54.47±0.70**	728.14 ± 3.46**	2.34 ± 0.126**

*Values represented as mean ± SEM of 6 animals, *p< 0.05, **p< 0.001, EEAI –Ethanolic leaf extract of Abutilon indicum*

CONCLUSION

The preliminary phytochemical investigations with ethanolic extract of *Abutilon indicum* (Linn.) Sweet leaves showed the presence of terpenes, phenol, essential oil especially eugenol which are confirmed by specific reactions.EEAI was screened for acute toxicity and was found to be non-toxic at the dose level of 200 mg/kg b.w. (p.o.) EEAI exhibited significant protection against water immersion plus restraint stress induced ulcer models. Histopathological studies of the stomach in stress induced ulcer models exhibit normal architecture of stomach tissue. EEAI was screened for *in vitro* antioxidant activity and it showed significant activity in the *in vitro* assays, like DPPH, reducing power, nitric oxide, and hydroxyl radical scavenging activity.

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