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Anti-ulcer activity of *Phyllanthus Niruri* and *Opuntia Ficus indica* plant extract

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Abstract

The present investigation carried out to study the anti-ulcer of opuntia ficus indica fruit extract and phyllanthus Niruri herb extract using two ulcer models Ethanol induced, and Pylorus ligation plus aspirin induced models.Effect of various doses of Opuntia ficus indica and Phyllanthus Niruri extract were studied on gastric secretion and gastric ulcers in Pylorus ligation plus aspirin induced and on ethanol- induced gastric mucosal injury in rats. The reduction in ulcer index in both the models along with reduction in volume and total acidity, proved the anti ulcer activity of opuntia ficus indica fruit extract and phyllanthus Niruri herb extract. Thus it can be concluded that both plant possesses anti-ulcer activity, which can be attributed to its anti-oxidant mechanism of action.

Key Words: Opuntia ficus indica and Phyllanthus Niruri, antiulcer, ethanol, pylorus ligation aspirin

Introduction

Peptic ulcer disease and its complications remain the cause of significant morbidity worldwide, representing a major burden for healthcare resources. Although potent anti-ulcer drugs are available, most of them produce several toxicities, thus emphasizing the need to search for new alternatives. As high as 80% of the world population depends on plant derived medicines for the first line of primary healthcare, reinforcing the theory that plant extracts can be good source of new durgs. [1] Peptic ulcer is one of the widespread gastrointestinal disorders. The various peptic ulcers are as follows:

- Duodenalulcer
- Gastriculcer
- NSAID inducedulcer
- Stressulcer

Management of pepticulcer:[2]

Peptic ulcer can be managed by following methods:

- By moderating gastric sourness, hyper motility and progressing ulcerremedial
- By inhibition of intricacies andrecurrence
- By treating the disease developed by *Helicobarterpylori*

Treatment:

- Withdrawal of the offending agents.
- Diet:
- Antacid and Anti-secretory drugs: eg Renitidine, omeprazole etc.
- Treating of *Helicobarter pylori* infection.
- Prevention of repetition:Ulcer repetition can be inhibited by combination of therapy like cimetidine and ranitidine.

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- **Surgery:** Surgery is preferable whenever
- Malignance is inferred in gastriculcer.
- A duodenal ulcer comes to be incurable and stubborn to suitable curative management.
- A complexity such as natural hindrance or prick isexisting.
- The patient bears from recurrent assaults of gastrointestinalhemorrhage.

Opuntia Ficus Indica, the Indian fig opuntia, fig opuntia, or prickly pear, is a species of cactus that has long been a domesticated crop plant grown in agriculture economies throughout arid and semiarid parts of the worlds and most commercially important cactus *PhyllanthusNiruri,* **was selected forthestudyaspotentialcorrosioninhibitor.*Phyllanthusniruri*is verycommonplantinIndiansystem,which has been used as anti-material, anti-bacterial and air purifier fromancienttime in Indian homes**

Material and Methods

Table: 1Detail of Plant Extract

Plant Name	Family	Species	Drug
Opuntia Ficus Indica	Cactaceae	*Opuntia Ficus Indica*	Fruits
PhyllanthusNiruri	Euphorbiaceae	Niruri	Herb

Animal:

The animal experimental protocol was approved by theDepartment of Pharmacy, Mansarovar University Bhopal**Institutional Animals Ethical Committee (IAEC), Deshpande Laboratories Pvt.Ltd. Bhopal(M.P.).**
CPCSEARegistration no**: 1582/PO/Re/11/CPCSEA.**

Pylorus ligatedulcer: Albino Wistar rats of either sex weighing between (150-200gms) were divided into various groups consist of six animals in each group.[3]

Group1 Normal Control
Group2 Control (Ethanol 1ml/200gm, p.o)
Group3 Standard (Omeprazole 20 mg/kg,p.o)
Group4 Ethanol extract of fruit of *Opuntia ficus-indica* (250 mg/kg, p.o)
Group5 Aqueous extract of fruit of *Opuntia ficus-indica* (250 mg/kg, p.o)
Group6 Ethanol extract of herb of *Phyllanthus niruri* (500 mg/kg, p.o)
Group7 Aqueous extract of herb of *Phyllanthus niruri* (500 mg/kg, p.o)

In this method albino rats were fasted in individual cages for 24 hours. Care was being taken to avoid coprophgy. Test compounds or standard drug or control vehicle was administered 30 minutes prior to pyloric ligation. The fasted rats were anaesthetized with anesthetic ether. An incision of 1cm long was made in the abdomen just below the sternum. The stomach was exposed. A thread was passed around the pyloric sphincter and a tight knot was applied. After pyloric ligation the abdomen wall was closed by putting the sutures. The skin was cleaned from any blood spots and bleeding. Collodion was applied over the wound. The rats were kept in a separate cage and allowed them to recover. After 4 hours of pyloric ligation the animals were sacrificed with excess of anesthetic ether. The abdomen was opened and the oesophageal end (cardiac end) of the stomach was tied. The entire stomach was cutted and removed from the body of the animal. A small cut was done to the pyloric region just above the knot and the contents of the stomach were collected in a graduated centrifuge tube. The gastric content was centrifuged at 1,000 rpm for 10 minutes and the volume was noted. The stomach was opened along the greater curvature and washed it slowly under the running tap water. Put it on the slide glass and observed under 10 x magnifications for ulcers. The ulcer scoring was done as per KulkarniSK[8]. [4]

0 = Normal coloured stomach
0.5 = Redcolouration
1 = Spotulcer

1.5 = Haemorrhagicstreaks
2 = Ulcers ≥ 3 but ≤5
3 = Ulcers >5
The mean ulcer score for each animal was expressed as ulcer index. The percentage protection was calculated using the formula.
% protection = 100 – ut/uc x 100
Where, ut = Ulcer index of treated group and uc = Ulcer index of controlgroup.

Determination of Free Acidity and TotalAcidity: 1ml of supernatant gastric content was pipette out and diluted it 10ml with distilled water. The pH of this solution was noted with the help of digital pH meter. The solution was titrated against 0.01*N* sodium hydroxide using Topfer's reagent as indicator until all traces of red colour disappeared and the colour of the solution turned to yellowish orange. The volume of sodium hydroxide added was noted. The volume of alkali was corresponded to free acidity. Then 2 to 3 drops of phenolphthalein was added and titration was continued until a red tinge reappears. Again the total volume of alkali added was noted. The volume was corresponded to totalacidity.[5]
Acidity was calculated by using the formula:
Acidity= Volume of NaOH x Normality of NaOH x 100mEq/L/100gm0.1

Estimation of Total Proteins: An alcoholic precipitate was prepared by adding 9.0 ml of 90% alcohol to 1 ml of gastric juice. To 0.1 ml of this protein mixture, 1 ml of 0.1N NaOH was added. 0.4 ml phenol reagent was added to 0.05 ml of the reaction mixture and was kept for ten minutes to complete the reaction. Absorbance was measured at 610 nm against blank in a spectrophotometer. The amount of protein present in the gastric juice was calculated from standard curve prepared with bovine albumin and was expressed in term of µg/ml of gastricjuice. [6]
Estimation of total carbohydrates: An alcoholic precipitate contains- ing the dissolved mucosubstances was prepared by adding 9 ml of 90% alcohol to 1 ml of gastric juice. The mixture was allowed to decant and the supernatant layer was discarded. The precipitate containing the mucosubstances was dissolved in 0.5 ml of 0.1N sodium hydroxide and 1.8 ml of 6N HCl was added. This mixture was boiled on a water bath, neutralised and diluted with distilled water to get a final volume of 4.5 ml. This solution was used for the estimation of carbohydrates like total hexoses, hexosamine, sialic acid and fucose asfollows.
Estimation of Fucose:
Reagents: Cysteine reagent: 600 mg of cysteine hydrochloride was dissolved in 20 ml of distilled water. It was stored in refrigerator until use. D (+) Fucose: Standard
Procedure: The blank and the sample tubes containing 0.4 ml of distilled water and 0.4 ml of hydrolysate was mixed carefully in a water bath with 1.8 ml sulphuric acid mixture. The reaction was allowed to take place for 3 minutes by heating on a water bath. After cooling the tubes 0.1 ml of cysteine reagent was added to the blank and to one of the tubes containing the hydrolysate (unknown) while cysteine reagent was not added to the test-tube containing the hydrolysate (unknown blank). The reaction was allowed to continue for 90 min. The absorbance was read spectrophotometrically at 396 and 430 nm using distilled water asblank.
The optical density for the fucose in the hydrolysate was calculated. The readings, which were taken at 396 and 430 nm, were noted and the difference calculated. Then the values without cysteine were subtracted from this and evaluated. Standard curve was prepared with D (+) – fucose. [7]

(OD396 – OD430) unknown – (OD396 – OD430) unknown blank
True optical density= ……
(OD396 – OD430) water blank the fucose content was expressed in

Estimation of Hexosamine:
Reagents: Acetyl acetone reagent: 0.3 ml acetyl acetone and 9.7 ml of 1.5N sodium carbonate (anhydrous) were mixed just before use. Ethanol: It was dehydrated.
Ehrlich's reagent: It was prepared by mixing 1.6 g of p-dimethyl amino benzaldehyde in 30 ml of 90 % ethanol and then adding 30 ml of Conc. HCl. The reagent was then stored in a refrigerator. D (+) Glucosamine hydrochloride was used asstandard.

Procedure: 0.5 ml of acetyl-acetone reagent was added to 0.5 ml of the hydrolysate fraction. The reaction was allowed to take place by heating for 20 min in a boiling water bath. The mixture was allowed to cool and 1.5 ml of 90% alcohol and 0.5 ml of Ehrlich's reagent was added. After 30 min the intensity of colour development was measured spectrophotometrically at 530 nm against blank. The amount of hexosamine present in the sample was estimated from the standard curve prepared by using D (+) glucosamine hydrochloride and concentration was expressed in µg/ml of gastricjuice.

Estimation of TotalHexoses:

Reagents: Orcinol reagent: 1.6 g of orcinol was dissolved in 100 ml of distilled water. This was prepared fresh before use. Sulphuric acid: 150 ml of conc. sulphuric acid was mixed with 100 ml of distilled water. D (+) Galactose- Mannose-StandardOrcinol-Sulphuric acid reagent: One volume of orcinol was mixedwith 7.5 ml of sulphuric acid. This was prepared fresh before use.

Procedure: The reaction mixture containing 0.4 ml of hydrolysate and 3.4 ml of orcinol reagent was heated for 15 min. in the boiling water bath. The tubes were cooled to room temperature and the colour developed was measured at 540 nm against the blank. The amount of hexoses present in the sample was determined from the standard curve of D (+) galactose - mannose and has been expressed in µg/ml of gastricjuice. [8]

Estimation of SialicAcid:

Reagents: Sodium arsenite: 0.5M sodium sulphate was prepared in 0.1N sulphuric acid and to 100 ml of this solution 10 g of sodium arsenite was added and stored in glass stoppered bottle.

Thiobarbituric acid: 0.5 M sodium sulphate was prepared and to 100 ml of this solution 600 mg of thiobarbituric acid was added and stored. Sialic acid- Standard.

Procedure: A reaction mixture containing 0.5 ml of the hydrolysate in $0.1NH_2SO_4$ and

0.2 ml of sodium periodate was mixed and allowed to stand for 20 min. 1 ml of sodium arsenite solution was added and mixed by shaking. 3 ml of thiobarbituric acid was added and the mixture was heated on a boiling water bath for 15 min. After cooling, 4.5 ml of cyclohexanone was added, thoroughly shaken and centrifuged. The pink colour formed in the supernatant layer was pipetted out and intensity of colour was measured spectrophotometrically at 550 nm. A standard curve was prepared using sialic acid and the amount of sialic acid present in the sample was determined expressed in µg/ml of gastricjuice. [9]

HistopathologicalEvaluation:

The stomachs of the all groups of animals were immersed in 10% formalin to study the histopathological changes. After the standard processing the wet ulcerated tissues were embedded in paraffin and cut into thick sections. Haematoxylin–eosin were used to stain the tissues and mounted with Canada balsam. Parameters used to study histopathological changes included shedding of gastric epithelium, gastric erosions, infiltration of neutrophils, oedema and inflammation. [10]

Results and Discussion

Pylorus ligated ulcer inrats

The anti ulcer activity of ethanol and aqueous extracts of *Opuntia ficus indica* and *Phyllanthus niruri* was studied at two dose levels (200 & 100 mg/kg) in pylorus ligated ulcerogenesis in rats.

Effect on Ulcer Index

The results indicated that *Opuntia ficus indica and Phyllanthus niruri* at dose levels of 200 mg/kg and 100 mg/kg significantly decreased the ulcer index ($p<0.01$) which was also evidenced by significant increase in percentage ulcer protection at both the dose levels. The percentage protection of ulcers in the treated groups at 200 and 100 mg/kg of ethanol extract of herb was found to be 47.86% and 77.78% respectively and aqueous extracts showed41.12% and 61.20% respectively. Omeprazole at 20 mg/kg showed a protection index of 88.89% (Table 2).

On the other hand, the percentage protection of ulcers in the treated groups at 250 and 500 mg/kg of ethanol extract of herb was found to be 55.6% and 70% respectively and aqueous extracts showed 46.66% and 65.60% respectively. (Table3).

Morphological appearances of ulcers induced by pylorus ligated ulcer in rats and treated with ethanol and aqueous extracts of *Opuntia ficus indica and*

Phyllanthus niruri herb and herb are shown in Plate 5.3.

Effect on Gastric Volume

Gastric volume in *Opuntia ficus indica and Phyllanthus niruri* treated groups indicated that there was no significant decrease in the volume of the gastric juice at 200 and 100 mg/kg. ($p<0.01$) (Table4).

Effect on pH of Gastric Juice

The ethanolic and aqueous extracts of herb and stem of *Opuntia ficus indica and Phyllanthus niruri* at 200 mg/kg and 100 mg/kg significantly increased the pH of gastric juice ($p<0.01$) and was comparable to the control and standard, Omeprazole at 20mg/kg (Table 4)

Table: 2Effect of *Opuntia ficus indica and Phyllanthus niruri* herb extracts on pylorus ligated rats

	Body weight	**Treatment**	**ULCER INDEX**						**Total score**	**Mean Ulcer Index ± SEM**	**% protec tion**
			Normal colourd stomach (0)	**Red colour-ation (0.5)**	**Spot ulcer (1.0)**	**Haemo-rrhagic streaks (1.5)**	**Ulcers ≥ 3 but ≤ 5 (2.0)**	**Ulcers >5 (3.0)**			
1	180	Normal	0	-	-	-	-	-	0	0±0	-
	170	Control	0	-	-	-	-	-	0		
	165		0	-	-	-	-	-	0		
	158		0	-	-	-	-	-	0		
	185		0	-	-	-	-	-	0		
	175		0	-	-	-	-	-	0		
2	170	Control	0	0.5	1.0	1.5	2.0	3.0	8.0	7.5±	0
	160	(Ulcerated)	0	0.5	1.0	1.5	2.0	-	5.0	0.500	
	155	Pylorous	0	0.5	1.0	1.5	2.0	3.0	8.0		
	165	Ligation	0	0.5	1.0	1.5	2.0	3.0	8.0		
	178		0	0.5	1.0	1.5	2.0	3.0	8.0		
	185		0	0.5	1.0	1.5	2.0	3.0	8.0		
3	155	Omeprazol	0	0.5	-	-	-	-	0.5	0.833±	88.89
	165	e(20mg/kg)	0	0.5	1.0	-	-	-	1.5	0.210	
	155		0	0.5	-	-	-	-	0.5	**	
	170		0	0.5	1.0	-	-	-	1.5		
	178		0	0.5	-	-	-	-	0.5		
	160		0	0.5	-	-	-	-	0.5		
4	178	Ethanol	0	0.5	1.0	-	2.0	-	3.5	3.91±	47.86
	155	Herb	0	0.5	1.0	1.5	2.0	-	5.0	0.351	
	170	Extract	0	0.5	1.0	1.5	-	-	3.0	**	
	175	(200mg/kg)	0	0.5	1.0	1.5	2.0	-	5.0		
	176		0	0.5	1.0	-	2.0	-	3.5		
	165		0	0.5	1.0	-	2.0	-	3.5		
5	180	Ethanol	0	0.5	1.0	-	-	-	1.5	1.666±	77.78
	175	Herb	0	0.5	1.0	-	-	-	1.5	0.333	
	165	Extract	0	0.5	-	-	-	-	0.5	**	
	170	(100mg/kg)	0	0.5	1.0	-	-	-	1.5		
	175		0	0.5	-	1.5	-	-	2.0		
	160		0	0.5	1.0	1.5	-	-	3.0		
6	155	Aqueous	0	0.5	1.0	1.5	2.0	-	5.0	4.416±	41.12
	158	Herb	0	0.5	1.0	1.5	2.0	-	5.0	0.374	
	165	Extract	0	0.5	1.0	1.5	-	-	3.0	**	
	170	(200mg/kg)	0	0.5	1.0	1.5	2.0	-	5.0		

	172		0	0.5	1.0	-	2.0	-	3.5		
	175		0	0.5	1.0	1.5	2.0	-	5.0		
7	182	Aqueous	0	0.5	1.0	1.5	-	-	3.0	2.91±	61.20
	175	Herb	0	0.5	1.0	1.5	-	-	3.0	0.153	
	160	Extract	0	0.5	-	-	2.0	-	2.5	**	
	168	(100mg/kg)	0	0.5	1.0	1.5	-	-	3.0		
	158		0	0.5	-	-	2.0	-	2.5		
	164		0	0.5	1.0	-	2.0	-	3.5		

Values are expressed as mean ± SEM (n=6) in each group.
*P <0.05, **P<0.01 compared with the control (ANOVA test)

Table: 3 Effect of *Opuntia ficus indica* fruit *and Phyllanthus niruri* herb extracts on pylorus ligated rats

	Body weight	**Treatment**	**ULCER INDEX**						**Total score**	**Mean Ulcer Index ± SEM**	**% protec tion**
			Normal colourd stomach (0)	**Red colour-ation (0.5)**	**Spot ulcer (1.0)**	**Haemo-rrhagic streaks (1.5)**	**Ulcers ≥ 3 but ≤ 5 (2.0)**	**Ulcers >5 (3.0)**			
1	180	Normal	0	-	-	-	-	-	0	0±0	-
	170	Control	0	-	-	-	-	-	0		
	165		0	-	-	-	-	-	0		
	158		0	-	-	-	-	-	0		
	185		0	-	-	-	-	-	0		
	175		0	-	-	-	-	-	0		
2	170	Control	0	0.5	1.0	1.5	2.0	3.0	8.0	7.5±	0
	160	(Ulcerated)	0	0.5	1.0	1.5	2.0	-	5.0	0.500	
	155	Pylorous	0	0.5	1.0	1.5	2.0	3.0	8.0		
	165	Ligation	0	0.5	1.0	1.5	2.0	3.0	8.0		
	178		0	0.5	1.0	1.5	2.0	3.0	8.0		
	185		0	0.5	1.0	1.5	2.0	3.0	8.0		
3	155	Omeprazol	0	0.5	-	-	-	-	0.5	0.833±	88.89
	165	e(20mg/kg)	0	0.5	1.0	-	-	-	1.5	0.210	
	155		0	0.5	-	-	-	-	0.5	**	
	170		0	0.5	1.0	-	-	-	1.5		
	178		0	0.5	-	-	-	-	0.5		
	160		0	0.5	-	-	-	-	0.5		
4	175	Ethanol	0	0.5	1.0	-	2.0	-	3.5	3.33±	55.6
	165	Herb	0	0.5	1.0	-	2.0	-	3.5	0.105	
	175	Extract	0	0.5	1.0	1.5	-	-	3.0	**	
	175	(200mg/kg)	0	0.5	1.0	1.5	-	-	3.0		
	176		0	0.5	1.0	-	2.0	-	3.5		
	170		0	0.5	1.0	-	2.0	-	3.5		
5	175	Ethanol	0	0.5	1.0	-	-	-	1.5	2.25±	70.0
	160	Herb	0	0.5	1.0	-	-	-	1.5	0.478	
	165	Extract	0	0.5	1.0	-	-	-	1.5	**	
	165	(100mg/kg)	0	0.5	1.0	-	-	-	1.5		
	168		0	0.5	-	1.5	2.0	-	4.0		

	155		0	0.5	1.0	-	2.0	-	3.5		
6	160 155 160 165 170 158	Aqueous Herb Extract (200mg/kg)	0 0 0 0 0 0	0.5 0.5 0.5 0.5 0.5 0.5	- 1.0 1.0 1.0 1.0 1.0	1.5 1.5 - 1.5 - 1.5	2.0 2.0 2.0 2.0 2.0 -	- - - - - -	4.0 5.0 3.5 5.0 3.5 3.0	4.0± 0.341 **	46.66
7	160 165 155 158 165 170	Aqueous Herb Extract (100mg/kg)	0 0 0 0 0 0	0.5 0.5 0.5 0.5 0.5 0.5	1.0 1.0 - 1.0 - 1.0	1.5 1.5 - 1.5 - -	- - - - 2.0 2.0	- - - - - -	3.0 3.0 0.5 3.0 2.5 3.5	2.58± 0.436 **	65.60

Values are expressed as mean ± SEM (n=6) in each group.
*P <0.05, **P<0.01compared with the control (ANOVA test)

Determination of free acidity and total acidity effect on free acidity and totalacidity

EstiofItion of gastric juice indicated that there was a significant (p<0.01) decrease in the free acidity and total acidity of the gastric juice in Animals treated with 200 mg/kg and 100 mg/kg of *Opuntia ficus indica and Phyllanthus niruri* and was comparable to that of Omeprazole (20 mg/kg) treated group (p<0.01) (Table 4

Table: 4 Effect of herb and herb extracts of *Opuntia ficus indica and Phyllanthus niruri* on antisecretory parameters of pylorus ligated rats

Gr. No.	Treatment	Does (mg/kg b.w.)	Gastric Volume (ml/100g)	pH	Free Acidity (mEq/l/100g)	Total Acidity (mEq/l/100g)
1	Normal Control	-	-	-	-	-
2	Control (Ulcerated)	-	4.86± 0.11	1.7 ± 0.096	73.88± 1.257	155.98± 2.344
3	Omeprazole	20 mg/kg	2.18± 0.079**	5.3 ± 0.141**	26.15± 0.761**	65.11± 0.326**
4	Ethanol Herb Extract	200 mg/kg	4.18± 0.079	2.71± 0.101**	63.58± 0.866**	131.80± 0.435**
5	Ethanol Herb Extract	100 mg/kg	3.08± 0.047**	4.05± 0.084**	46.71± 0.819**	91.30± 0.447**
6	Aqueous Herb Extract	200 mg/kg	3.85± 0.111*	2.85± 0.105**	61± 0.957**	124.46± 0.566**
7	Aqueous Herb Extract	100 mg/kg	3.11± 0.083**	3.08± 0.101**	49.33± 0.539**	98.18± 0.440**

8	hanol Herb Extract	250 mg/kg	4.01± 0.070	2.88± 0.047**	58.83± 0.398**	112.25± 0.863**
9	hanol Herb Extract	500 mg/kg	3.25± 0.18**	3.53± 0.076**	49.7± 0.364**	90.08± 0.426**
10	Aqueous Herb Extract	250 mg/kg	3.55± 0.042 **	2.9± 0.073**	56.96± 0.683**	106.93± 0.560**
11	Aqueous Herb Extract	500 mg/kg	2.9 ± 0.051**	3.11± 0.166**	48.5± 0.476**	95.23± 0.398**

Values are expressed as mean ± SEM (n=6) in each group.
**P* <0.05, ***P*<0.01 compared with the control (ANOVA test)

Effect on totalproteins

The ethanol and aqueous extracts of herb and herb of *Opuntia ficus indica and Phyllanthus niruri* indicated the results that the total protein content was significantly decreased (p<0.01), in the treated group at 200 and 100 mg/kg when compared to the control (Table 5)

Table: 5 Effect of *Opuntia ficus indica and Phyllanthus niruri* herb and herb extracts on total proteins and C/P of pylorus ligated rats

Gr. No.	Treatment	Does (mg/kg b.w.)	Total Proteins (µg/ml)	C/P
1	Ulcerative Control	-	487.16± 0.792	0.83
2	Omeprazole	20 mg/kg	244.81± 1.249**	4.63
3	Ethanol Herb Extract	200 mg/kg	412.66±0.802**	1.52
4	Ethanol Herb Extract	100 mg/kg	278.82± 0.666**	2.97
5	Aqueous Herb Extract	200 mg/kg	431.16±1.194**	1.43
6	Aqueous Herb Extract	100 mg/kg	310.66±0.560**	2.44
7	Ethanol Herb Extract	250 mg/kg	422.16±0.703**	1.57
8	Ethanol Herb Extract	500 mg/kg	301.66±0.666**	2.55
9	Aqueous Herb Extract	250 mg/kg	435.33±0.802**	1.55
10	Aqueous Herb Extract	500 mg/kg	292.65±0.792**	2.63

Values are expressed as mean ± SEM (n=6) in each group.
*P <0.05, **P<0.01 compared with the control (ANOVA test)

Effect on totalcarbohydrates

Carbohydrates content of *Opuntia ficus indica and Phyllanthus niruri* (Linn.) treated group indicated that there was a significant increase (p<0.01) in the total carbohydrate content at 250 and 500 mg/kg treated group and standard group was comparable to the control group. (Table 6)

Table: 6 Effect of *Opuntia ficus indica and Phyllanthus niruri* herb and herb extracts on total carbohy- drates of pylorus ligated rats

Gr.No.	Treatment	Does (mg/kgb.w.)	Total Carbohydrates (µg/ml)			
			Total Hexose	Hexosamine	Fucose	Sialic acid
1	Control (Ulcerated)	-	153.5 ±0.562	166.5 ±0.562	69.66 ±0.557	18.83 ±0.703
2	Omeprazole	20 mg/kg	498.83 ±1.013**	428.33 ±0.802**	155.66 ±0.666**	52.16 ±0.542**
3	Ethanol Herb Extract	200 mg/kg	286.16 ±0.600**	222.16 ±0.945**	81.83 ±0.600**	38.83 ±0.703**
4	Ethanol Herb Extract	100 mg/kg	370.45 ±0.392**	320.66 ±0.402**	92.64 ±0.569**	44.36 ±1.636**
5	Aqueous Herb Extract	200 mg/kg	274.32 ±1.021**	227.54 ±0.365**	74.66 ±0.687**	40.22 ±0.721**
6	Aqueous Herb Extract	100 mg/kg	332.83 ±1.016**	298.36 ±0.0324**	92.65 ±0.709**	36.22 ±0.333**
7	Ethanol Herb Extract	250 mg/kg	280.22 ±0.666**	270.35 ±0.065**	79.55 ±0.365**	36.87 ±0.332**

8	Ethanol Herb Extract	500 mg/kg	335.21 ±0.663**	302.66 ±0.982**	90.66 ±0.709**	40.88 ±0.630**
9	Aqueous Herb Extract	250 mg/kg	287.66 ±0.323**	279.65 ±0.221**	70.36 ±0.666**	39.83 ±0.731**
10	Aqueous Herb Extract	500 mg/kg	350.55 ±0.362**	301.66 ±0.695**	87.25 ±0.321**	32.64 ±1.402**

Values are expressed as mean ± SEM (n=6) in each group.
*P <0.05, **P<0.01 compared with the control (ANOVA test)

Histopathologicalstudies

The control group of rats treated with pyorus lgation induced ulcer showed histopathological changes of the gastric mucosa characterized by loss of glandular architecture, oedema and erosions of the epithelial layer, evident oedema, congestion and infiltration by inflammatory cells. The rats treated with the ethanol and aqueous extracts at 200 and 100 mg/kg, b.w showed minimum ulceration and oedema but gastric epithelium was not intact. However, at a dose of 200 and 100mg/kg ethanol and aqueous extracts of fruit and herb of *Opuntia ficus indica and Phyllanthus niruri*, the rats showed significant regenerative changes indicating healing

Figure: 1 Histopathological studies of the antiulcer activity of *Opuntia ficus indica and Phyllanthus niruri* by pylorus ligated ulcer in ratsa Section of the stomach of control group of pylorus ligated ulcer in rats.

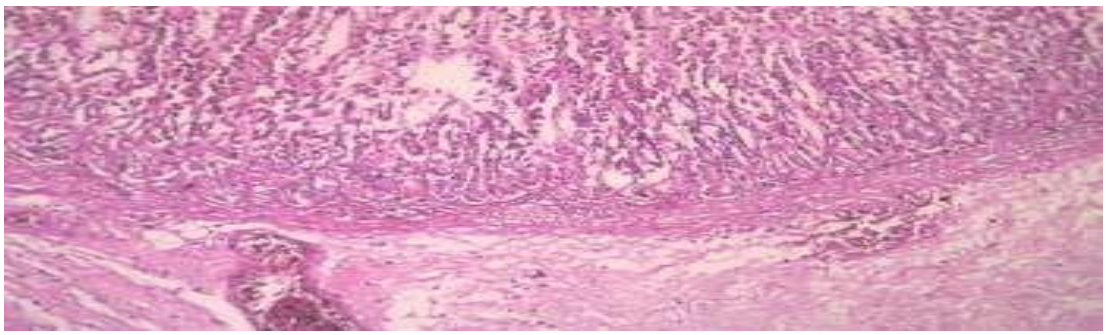

Figure: 2 Section of the stomach of Omeprazole (20 mg/kg b.w) treated group of pylorus ligated ulcer in rats.

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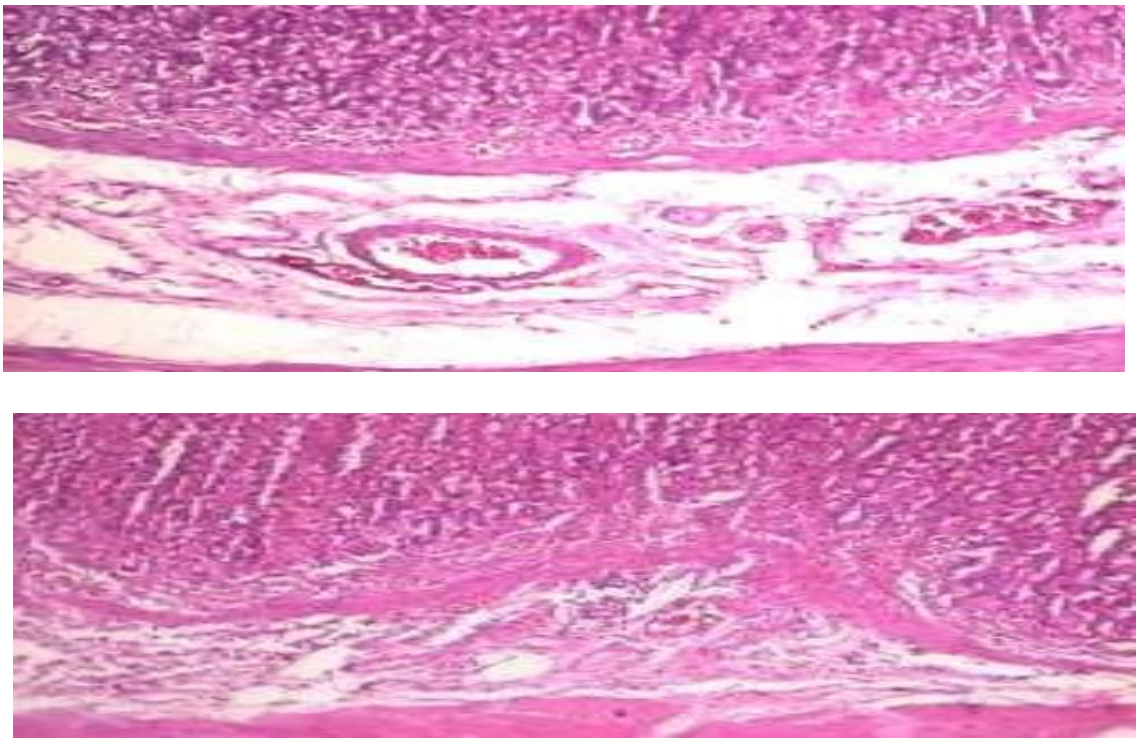

Figure: 3Section of the stomach of ethanol extract of *Opuntia ficus indica and Phyllanthus niruri* herb (200 mg/kg b.w) treated group of pylorus ligated ulcer in rats.

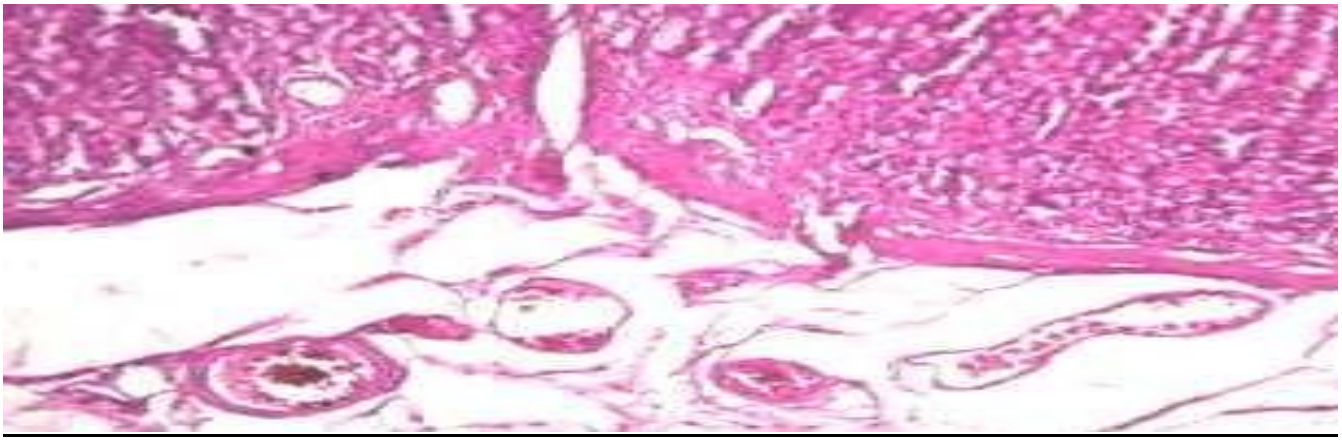

Figure: 4 Section of the stomach of aqueous extract of *Opuntia ficus indica and Phyllanthus niruri* herb (100 mg/kg b.w) treated group of pylorus ligated ulcer in rats.

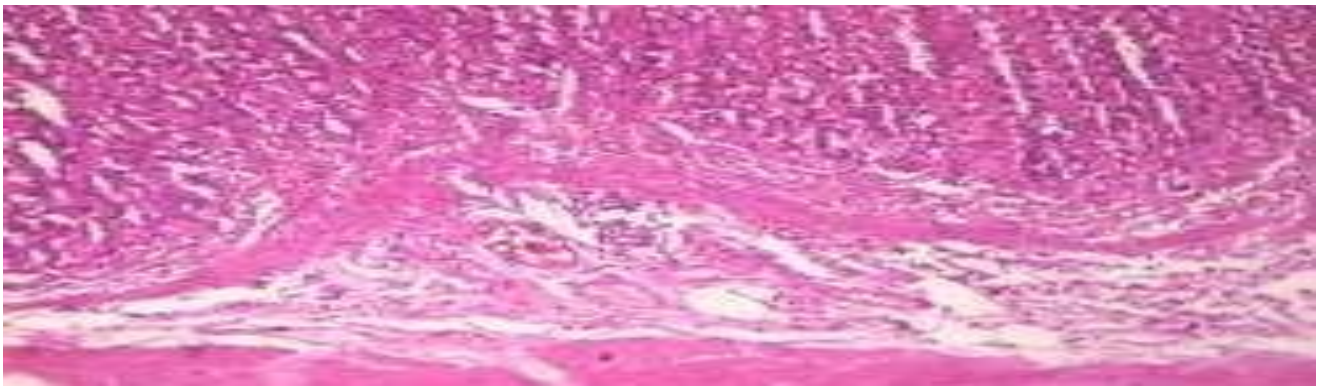

Figure: 5Section of the Stomach of ethanol extract of *Opuntia ficus indica and Phyllanthus niruri* herb (200 and 100mg/kg b.w) treated group of pylorus ligated ulcer in rats.

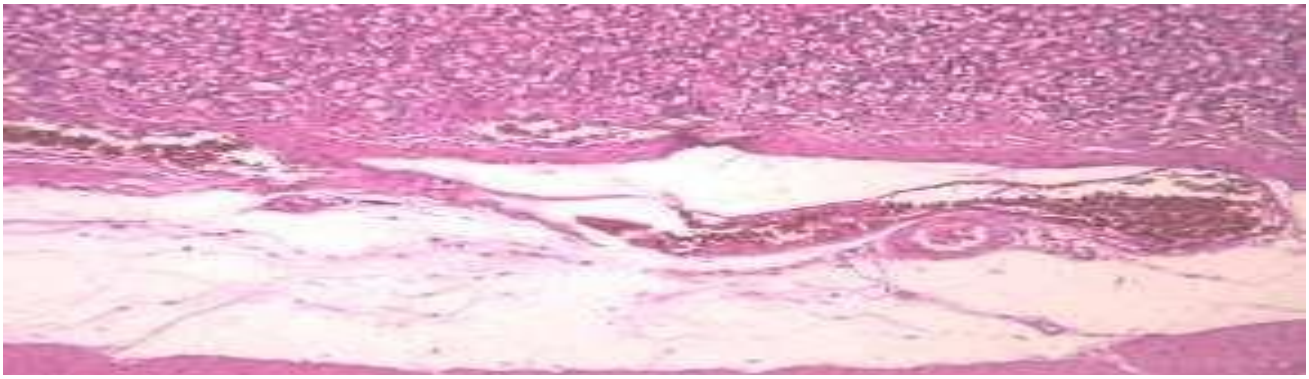

Figure: 6 Section of the stomach of aqueous extract of fruit of*Opuntia ficus indica and Phyllanthus niruri*herb (200 and 100mg/kg b.w) treated group of pylorus ligated ulcer in rats.

Conclusion

From the above results it can be inferred that the ethanol extracts of *Phyllanthus Niruri and Opuntia ficus-indica*displayed significant anti-ulcer activity. The extracts impart anti-ulcer activity due to presence of polyphenol and flavonoids component. These phytoconstituents scavenge the free radical produce by pylorus ligation and reduced the gastric mucosal damage. Moreover, it also protects the stomach from mucosal injury by inhibiting the succession of gastric ulcers. Accordingly, the anti-ulcer activity of the extracts is referred to the impressive antioxidant properties of theextracts.

Additionally, it is also documented that terpenoids heal the ulcer present in stomach, due to the activation of cellular protection, reduction of mucosal prostaglandins metabolism, cytoprotective action and reduction of gastric vascular permeability. On the basis of phytochemical investigation we also referred that extract contributes antiulcer activity due to terpenoids present in extracts.

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