



Pharmacognostical Evaluation and Screening of *Terminalia paniculata* roth Bark for Mucoprotective activity

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Abstract: In the present investigation, the pharmacognostical evaluation was carried out to study its morphology and microscopy of *Terminalia paniculata* Roth. bark and its powder characters, followed by determination of proximate values. Hydro alcoholic bark extract of *Terminalia paniculata* Roth. (HBTP) was subjected to qualitative phytochemical screening by well established methods to confirm the presence of important primary and secondary metabolites. The total content of polyphenols in the extract was evaluated, spectrophotometrically as per the Ciocalteu model, The overall flavanoid contents of extract were determined by Aluminium chloride colorimetric assay. The gastric antiulcer activity of hydro alcoholic extract of bark *Termenalia paneculata* Roth. was performed by employing ulcerogenic models like ethanol-induced mucosal damage in rats. HBTP (200mg and 400 mg/kg) was administered for 15 days and 16th day 1ml/200gm absolute ethanol was administered orally. Sucralfate was used as standard drug (100mg/kg). After 4 h of administration of absolute ethanol, animals was sacrificed and stomach was incisioned for determination of severity of ulcer. The parameters like index of ulcer, volume of gastric, pH, total acid content and gastric mucin level were measured followed by their histopathological studies. The hydroalcoholic bark extract of plant (400 mg/kg, per.oral) revealed significant ($p < 0.001$) antiulcer property.

Keywords: *Terminalia paniculata* Roth. Bark, pharmacognostic, phenolic content, flavonoid content, mucoprotective, Histopathology.

INTRODUCTION

“Peptic ulcer” often reveals to the wounds or sores that will appear on the inner surface of certain parts of the g.i. tract viz. stomach and upper small intestine, secondary to the increased gastric acid or reduce protectivity of gastric mucosa or bacterial infections[1]. Based on their proper location, two terms are used to indicate peptic ulcers. First, gastric ulcers denote ulcerations on the lining of the stomach, and second, duodenal ulcers indicates ulcerations on the wall of duodenal [2].

Other prominent, non-pathogenic route to gastric ulceration is the increase in administration of non-steroidal anti-inflammatory drugs (NSAIDs), more commonly known as the “relievers of pain”. Life long administration of these drugs reduces bicarbonate and mucous synthesis, mucous blood circulation, platelet of aggregation that can cause depletion of the protective mucus layer and expose the gastric wall to acid-induced damage and irritation [3]. Presently, the treatment mode for peptic ulcer is proton pump inhibitors which are indicated by omeprazole (along with its pure enantiomer

like sucralfate, pantoprazole, rabeprazole and lansoprazole. Other older option that is less indicated in present days as well as less effective, used rarely, administration of H_2 -receptor antagonists including cimetidine, ranitidine, famotidine, nizatidine and roxatidine. At present, the use of natural sources for the protection and ailment of peptic ulcer is not casual and is often recommended over new medicines. In olden times, many research institutes around the globe have made use of common plant parts to reduce gastric disorders[4]. A genus of *Termenalia* which are medium to large trees of Combretaceae family with about 250 species are there in the world. Various species are widely included in many ayurvedic systems. The genus “*Termenalia*” is obtained from Latin term “*Terminus*”, which means the leaves are located at the tip of the branch[5]. The *Termenalia* plants bark has cracks and branches tucked into many of layers. Many *Termenalia* plants’ leaves are big, leathery and solitary green white flowers. The fruits are yellow, dark, red or black in colour, usually angular and winged. Some fruits are consumable, tonic and possess therapeutic properties[6].



Fig-5.1 Photographs showing aerial parts of *Terminalia paniculata* Roth

Members of genus *Terminalia* have been used for a wide range of therapeutic reasons by traditional healers from a broad range of ethno - cultural groups. Its external application helps in wound healing and early reunion of Fractures[7] . It contain. fourteen per cent of tanins, have a nucleus of pyrogallol in the molecule, other than a catechol nucleus with gallic acid [8] . β -sitosterol were separated and recognised from the pet. ether extract of the bark[9]. In HPLC and spectroscopic detection, presence of compound marker such as gallic acids, ellagic acids, catechin and epicatechins[10]. Galloylarjunolic acid, 3,3'-di-O-Methyl ellagic acids, Arjunolic acid, Betulenolic acid, 23-O-galloylarjunic acid, Catechin Arjunetin, Oleonolic acid, Ursolic Acids, Arjunogenin Arjunolic acids, Betulenolic acids, Chebulegic acid, Orientine, Isovitexen, Vitexen, Quercetin, Kaempferol-3-O- β -D-rutinosides, Rutin, Quercetin-3,4'-di-O-glucoside[11], The glucoside of dimethyl ellagic acid has been separated from the heartwood of *Terminalia paniculata* together with β -sitosterols and novel triterpene carboxylic acids[12]. Pharmacologically plant were reported for anti-obesity and Antidiabetic activity[13,14] Antimicrobial activity[15] Antioxidant activity[16].

Till now there are no available reports on the pharmacognostical studies and mucoprotective activity of *Terminalia paniculata* Roth. bark extracts. Therefore the present study was undertaken to evaluate the pharmacognostical parameters and mucoprotective property of *Terminalia paniculata* Roth. hydroalcoholic bark extracts in ethanol-administered mucosal damage in albino rats.

Materials and methods

Collection and authentication of the plant material:

Bark of *Terminalia paniculata* Roth. were collected from Western Ghats. the plant Authentication was carried out by taxonomist by Dr. S. S. Hebbar, Botanist, Government P.U.C. College, Dharwad. A specimen of herbarium of the plant was kept in Pharmacognosy Department, SET's College of Pharmacy, Dharwad, Karnataka, Herbarium accession No SETCPD/Ph.cog/herb/75/2022.

Experimental Animals:

Wistar rats of Male weighing 150-200 g were used for the study. The animals were kept under controlled conditions of temperature ($22 \pm 2^\circ\text{C}$), humidity ($50 \pm 5\%$) and 12-h light/dark

cycles. They were given standard diet and water, *ad libitum*. All the experiment carried out were approved by the Institutional Animal Ethical Committee (IAEC) of SET's College of Pharmacy, Dharwad, Karnataka (841/PO/Bt/CPCSEA Date: 15.02.2017) as per the guidelines of Committee for the Purpose for Control and Supervision of Experiments on Animals (CPCSEA), Government of India. SET/CP/IEAC/179/04.Dated 25.03.2023 .

Methods

Pharmacognostical evaluation:

The pharmacognostic studies helps in identification of plant and their species.

Macroscopical Features:

Macroscopical features and external characters such as colour; odour, taste, shape and size, texture was observed and analysed botanically.

Study of Microscopical properties:

Transvers section and Powder microscopy of bark of *Terminalia paniculata*:

A few drops of chloral hydrate solution was added to a thin section of bark on a slide, covered with a glass slip and heated gently over a spirit lamp. Rapid boiling was avoided. The slide was examined under the microscope. When the process is completed a drop of glycerol solution were added which will prevent crystallization process of the mounting agent on cooling. The excess reagent were removed by piece of filter paper and section was observed for various cellular characters under 10x, 45x and 100x lens. The same procedure was carried out for powder microscopy to view the important diagnostic characters.

Evaluation of Physical constant

The standardization of the *Terminalia paniculata* Roth. bark was carried out by evaluating physical-chemical paradigms like percentage of moisture content, extractive and ash content. The following parameters were carried out as per well established methods.

1. Extractive content:

a. Alcohol soluble content:

b. Water soluble content:

2. Percentage of Moisture:

3. content of ash:

a. Ash total value:

b. Ash Acid-insoluble:

c. Ash Water-soluble:

d. Ash Sulphated: [16].

Preparation of Hydro alcoholic bark extract:

Terminalia paniculata Roth. bark powder was extracted with 70% ethanol in hot continuous extraction by Soxhlet method. The extracts was concentrated in a rotary flash and residue was dried in a desiccator over anhydrous reagent to give hydro alcoholic concentrate [HBTP]. The % of hydro alcoholic concentrate was determined.

PHYTOCHEMICAL ANALYSIS:

The phytochemical evaluation was carried out for hydro ethanolic bark extract of *Terminalia paniculata* Roth. The tests were analysed by well established parameters[17].

First Group	Normal group
Second Group	Positive group (1ml/200gm (99%) absolute ethanol ,orally)
Third Group	Standard drug (Sucralfate 100 mg/kg, orally)
Fourth Group	HBTP (200mg/kg) + Ethanol
Fifth Group	HBTP (400mg/kg)+ Ethanol

All the animals were administered as per the grouping mentioned above for 15 days. After the last dose, animals are not given the food for 24 hours before administration of absolute ethanol. Each rat was administered with the 1ml/200gm absolute ethanol orally. The rats were sacrificed after 4 h of ethanol treatment as per the CPCSEA guideline and a incision was made at midline and the stomach is removed, then the stomach was opened along with greater of curvature, stretched gently by pinning on a cork board, afterwords the gastric mucosa was analysed by naked eyes and magnifying lens[20].

Morphological Analysis of the Stomach:

The stomach was incisioned along with the larger curvature, dipped in normal saline to remove gastric residues, and analysed under 10x magnifying lens to view the ulcer formation. The overall number of ulcers were tallied and given following grades (0 = no ulcer, 0.5 = red coloration, 1 = spot ulcers, 2 = deep ulcers, and 3 = perforations). then, the ulcer of index (UI) and percentage ulcer inhibition (%UI) were analysed using the following formula:

$$\text{Ulcer Index} = \frac{\text{Sum of lesion areas}}{\text{Total Stomach areas}} \times 100$$

Where UI = ulcer of index; UN = average numbers of ulcer for each animal; US = average of severity score, and UP = percentage of ulcer of animals.

Percentage of ulcer protection:

The percentage of ulcer protection was calculated using following formula:

$$\text{Percentage of Ulcer Protection} = \frac{\text{MUI of control} - \text{MUI of test}}{\text{MUI of control}} \times 100$$

Where, MUI= Mean Ulcer Index

QUANTITATIVE EVALUATION OF POLYPHENOLIC COMPOUNDS:

Determination of total content of phenols and content of flavonoids was carriedout by standard methods:[18].

Ethanol-induced mucosal damage:

The Mucoprotective role of hydro alcoholic bark concentrate of *Terminalia paniculata* Roth. were analysed in ethanol-administered mucosal damage in rats. Based on literature survey two doses i.e 200mg/kg and 400mg/kg of hydro ethanolic bark extract of *Terminalia paniculata* Roth. were selected for evaluation of Mucoprotective activity[19].

Experimental design:

The Mucoprotective activity of hydro alcoholic bark extract of *Terminalia paniculata* Roth (HBTP) was determined in ethanol-induced mucosal damage in rats. The albino wistar rats weighing 150-200g of either sex were divided into following groups consisting of six animals each.

Analysis of Volume of Gastric and PH:

The contents of stomach were made empty from measuring tubes, centrifuged for 10min at a 1000rpm, afterwards volume of the supernatant was recorded then, 1ml of gastric juice was removed from supernatant, mixed with 1milliliter of distill water, and the pH of the solution were analysed using a PH meter.

$$\text{Acidity} = \frac{\text{Vol. of NaOH} \times \text{Normality} \times 100 \text{ mEq/L}}{0.1}$$

Analysis of Gastric Content of Mucus:

The glandular sections from the stomach were desected and weighed. For 2 h, each section was dipped in 10 mL of 0.1% Alician blue of 8GX solution (in 0.16 Molar sucrose solution, which is buffered with 0.05 M sodium acetate, adjusted to pH 5.8). Then residual dye was made cleared by washing twice with 0.25 Molar sucrose solution for 15 and 45 min. Dye were chelated with mucus on the gastric wall, then separated using 20 mL of 0.05 M of magnesium chloride with occasional stirring. Then, 5 mL volume of blue extract was shaken for 2 h for every 30 min with an equal amounts of diethyl ether. Using a UV-visible spectrophotometer at 580 nm, the mixture was centrifuged about 15 min at 3000 rpm, and the absorbance of aqueous layer were analysed and recorded. Concentration of Alician blue extracted/g (net) of gland tissue were then compared with the alician blue standard curve[21].

STATISTICAL ANALYSIS:

The data were expressed as Mean $\pm$ S.E.M. Statistical comparisons were performed by one-way ANOVA followed by Tukey's post - test using Graph Pad Prism version 5.0, USA.

Results

PHARMACOGNOSTICAL EVALUATION:

Surface Features:

The bark is thick and woody outer surface brown with numerous pits. as shown in (Figure: 5.1-A) The inner surface is dark brown with vertical thin, parallel shallow depression. as show in (Figure 5.1-B)

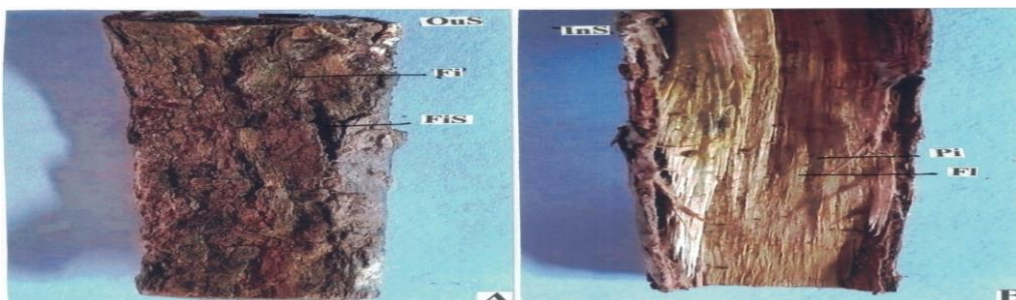


Figure 5.1(A,B) Photographs showing Surface Features of *Terminalia paniculata* bark

Ous: Outer surface, Ins: Inner surface, Pi: Pits, FI: Fibre, Fis: Fissured

Microscopical Characteristics:

Transverse Section of *Terminalia paniculata*, Roth. bark:

Anatomy of the Bark:

The bark is irregularly broken on the surface forming thick ridges and periderm as show in (Figure 5.2-1&2). The periderm consist of horizontal thick layers of periderm which includes phellem followed by inner phelloderm. phellem layer occur in a few successive layers of phellem cells. Where the cells are heterocellular, the phellem includes several horizontal oblong narrow layers of lignified cells, which are called rhytidome as show in (Figure 5.4-1&2).

When viewed under polarised light the rhytidome cells appear a bright white due to presence of lignin in the cell wall as show in (Figure: 5.4-2). In between the rhytidome cells are seen radially oblong wide. Thin walled cells which are suberised by dead phellem cells as show in (Figure 5.4-1 &2).

Inner to the periderm occurs wide parenchymatous cortical tissue. The cortical cells are small, angular with dense tannin contents as show in (Figure. 5.3). The inner most region includes thin cylinder of secondary phloem as show in (Figure. 5.3). Secondary phloem is differentiated in to outer collapsed phloem where the cells are densely filled with tannin and calcium oxalate druses as show in (Figure 5.5-1).

Inner to the collapsed phloem occurs non collapsed phloem as show in (Figure 5.5-1 & 2). The non collapsed phloem includes phloem parenchyma, sieve elements, and companion cells as show in (Figure. 5.5-2). All the cells are wide and thin walled. The sieve elements are circular possessing. narrow rectangular Companion cells attached on the lateral walls (Figure. 5.5-2). These sieve elements are 60µm in diameter. The non collapsed phloem cells do not possess druses and tannin.

TLS of Phloem:

In the TLS view, the sieve elements appear as thin narrow vertically oblong sieve elements. The sieve plate is oblique and thin walled as show in (Figure 5.6-1 & 5.7-1, 2). The phloem rays are thin narrow and uniseriate. The rays possess vertically oblong spindle shaped and possess dense vertical layers of large spherical calcium oxalate druses as show in (Figure 5.6-.2).

Apart from the phloem rays there are vertical layers of parenchyma cells. The oxile parenchyma cells are vertically elongated arranged one below the other. Occasionally there are biseriate ray as show in (Figure 5.7-1&2)

The phloem rays have central part with short rectangular cells which are called procumbent cells, at the ends of the ray, cells are vertically elongated and triangular in shaped. These cells are called upright cells. The ray consisting of upright and procumbent are called hetero cellular this cells having only one type of cell (only upright cell or procumbent cell) and RLS were depicted in Fig-5.8-5.9 (1 and 2).

Powder microscopy of *Terminalia paniculata*, Roth, bark

Powder Preparation of the bark has rarely trichome as fibre with blend end and wide lumen as show in (Figure: 5.10-1). These are also typical wide fibre with wide central back and tapering ends. The cell lumen is a wide and the cell wall is thin as show in (Figure: 5.10-2&3). The wide fibre are 460µm to 560µm long and 30µm wide as show in (Figure: 5.11-1) also contains smallest pieces of parenchyma cells. The pieces are polygonal with thick walled cells.

Occasionally rich tracheids are also seen in the powder, Tracheids are long, wide, and cylindrical in shape. Do not have end wall perforation and they have numerous vertical rays of border pits 50µm as shown in (Figure: 5.11-2)

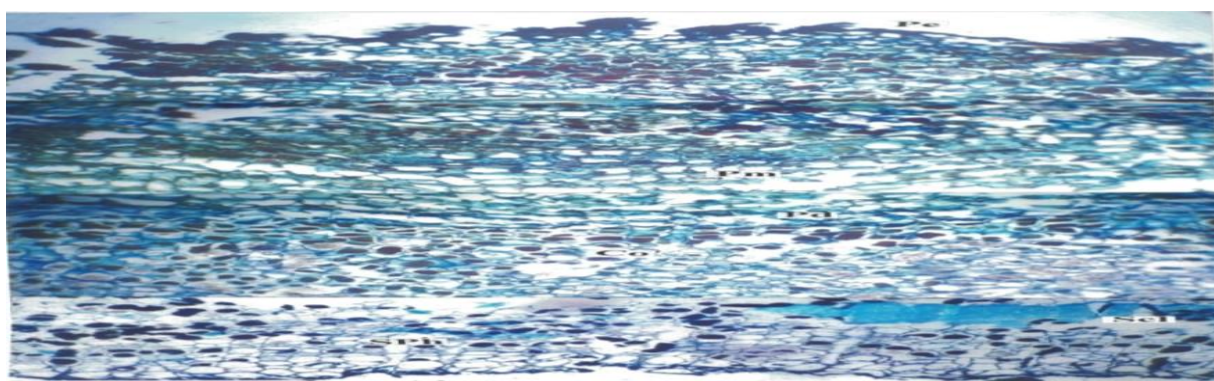


Figure 5.2 T.S. of bark showing Three segments of periderm, Cortex and secondary Phloem Co-Cortex, Pe-Periderm, Pd-Phelloderm, Pm-Phellen, Scl-Sclerides, Sph- Secondary Phloem

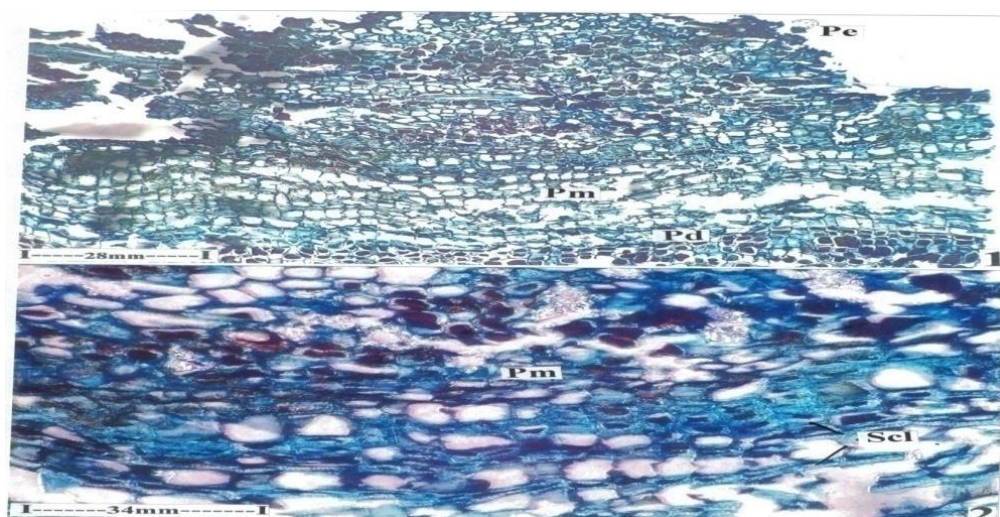


Figure: 5.3 1. T.S. of Periderm with broken outer tissue and heterogenous phellem and inner parenchymatous Phelloderm.
2. Phellem with thin walled suberised cell and thick wall lignified Phelloid cells. Pe – Periderm, Pd – Phelloderm, Pm-Phellem, Scl-Sclerides.

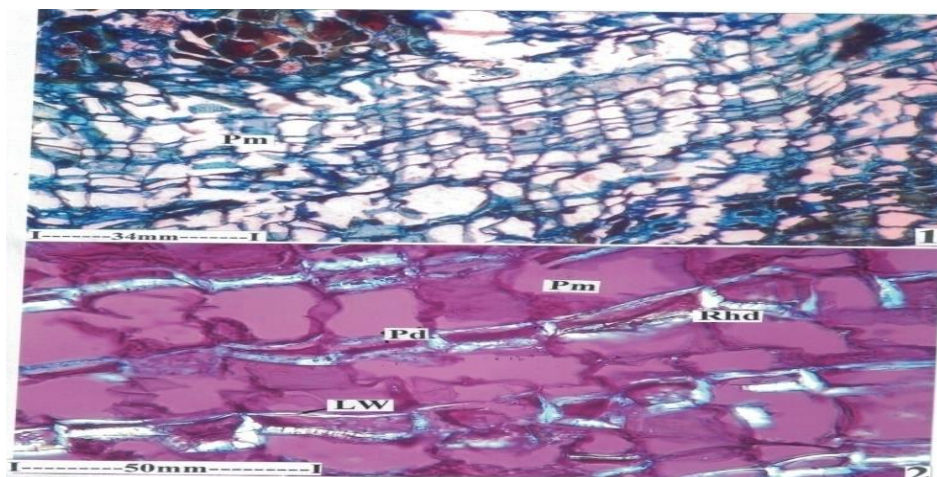


Figure: 5.4

1 Phellem tissue showing wide thin walled suberised cell wall, and alternating with horizontal layer to rectangular lignified Phelloid.

2: Phellem and Phellod cells enlarged under the polarised light.

Pm- Phellem, Pd – Phelloderm, LW -Lignified Walls, Rhd- Rhytidome

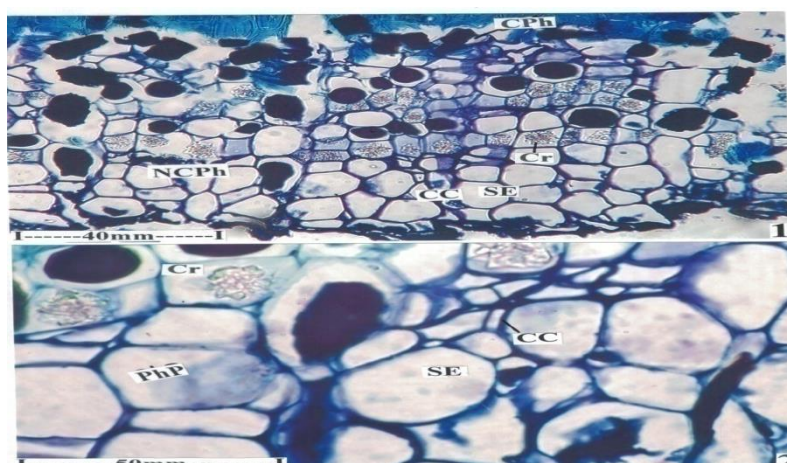


Figure: 5.5

1. Secondary phloem enlarged showing outer collapsed sieve element and lines non-collapsed sieve element. Most of the cell have block circular mass of tannin.

2. Non-collapsed phloem element enlarged, showing sieve elements and companion cell.

CC-Companion Cell, Cr-Crystal, NCPH-Non Collapsed Phloem,

PhP-Phloem Parenchyma, SE-Sieve Element

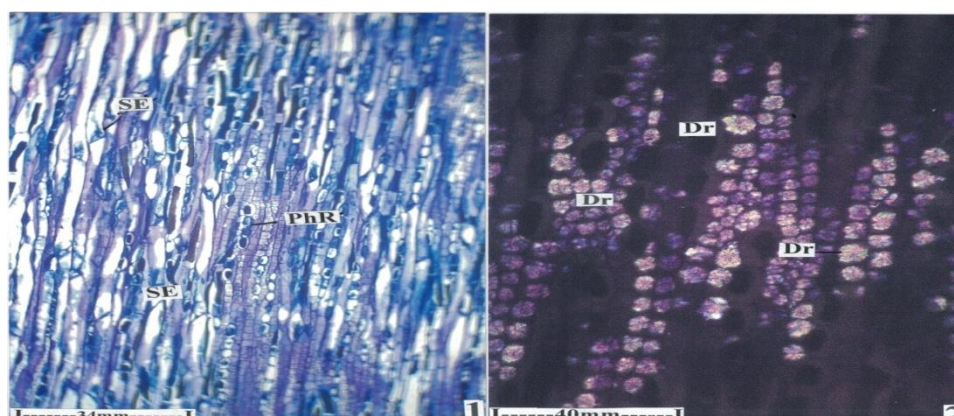


Figure: 5.6.

1. TLS view of Secondary Phloem having phloem ray and in Sieve element Sieve element,

2. TLS of phloem Showing numerous vertical straight line of circular a spherical druses.

Dr–Druces, PhR-Phloem Ray, SE-Sieve Element.

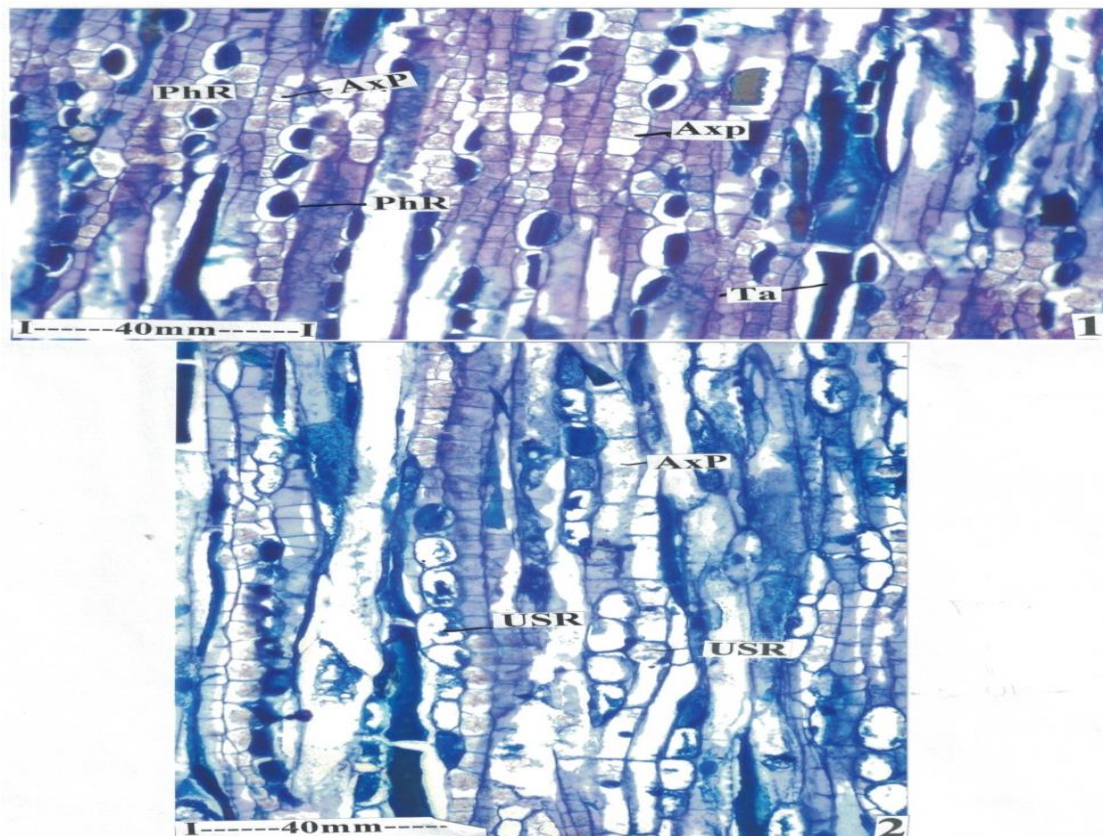


Figure: 5.7 1. TLS of phloem ray and oxile parenchymatous cell
2. TLS of Phloem axile parenchyma and uniseriate ray

AxP- Axile Parenchyma, PhR- Phloem Ray, Ta- Tannin, USR- Uniseriate Ray

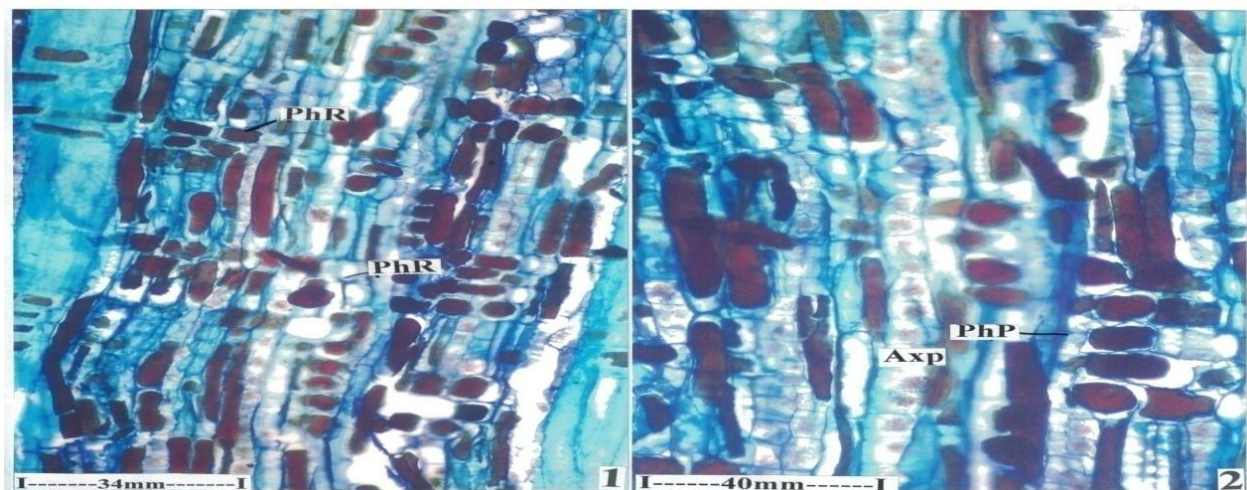
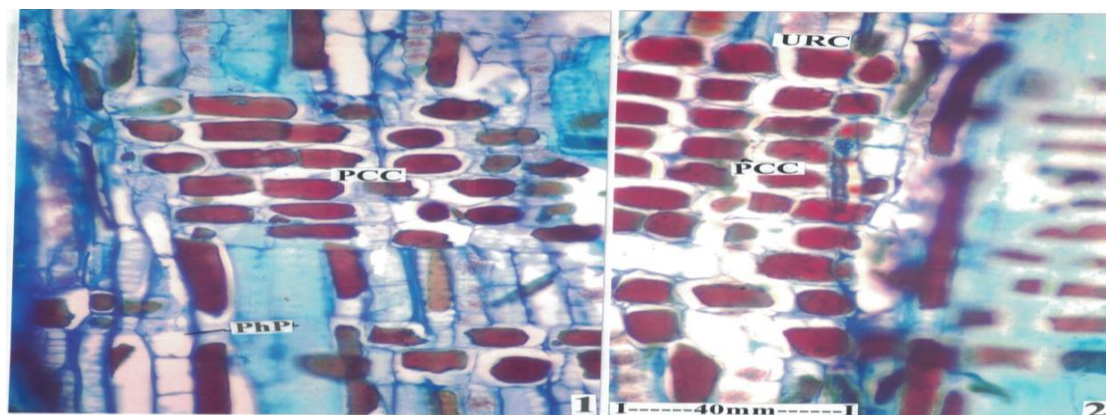


Figure: 5.8 1 & 2 RLS of Phloem Ray. The ray have horizontal rectangular parenchymatous cells are procumbent cells and marginal vertical axile Parenchyma cells is called upright cells.

Axp- Axile Parenchyma, PhP- Phloem Parenchyma, PhR-Phloem Ray



Figure; 5.9

1 & 2 In enlarged View the procumbent cells appear horizontally rectangular and upper and lower erect rectangular cells are upright cells and all the cells have dark brown tannin

PCC-Procumbent Cell, URC - Upright Cell, PhP-Phloem Parenchyma

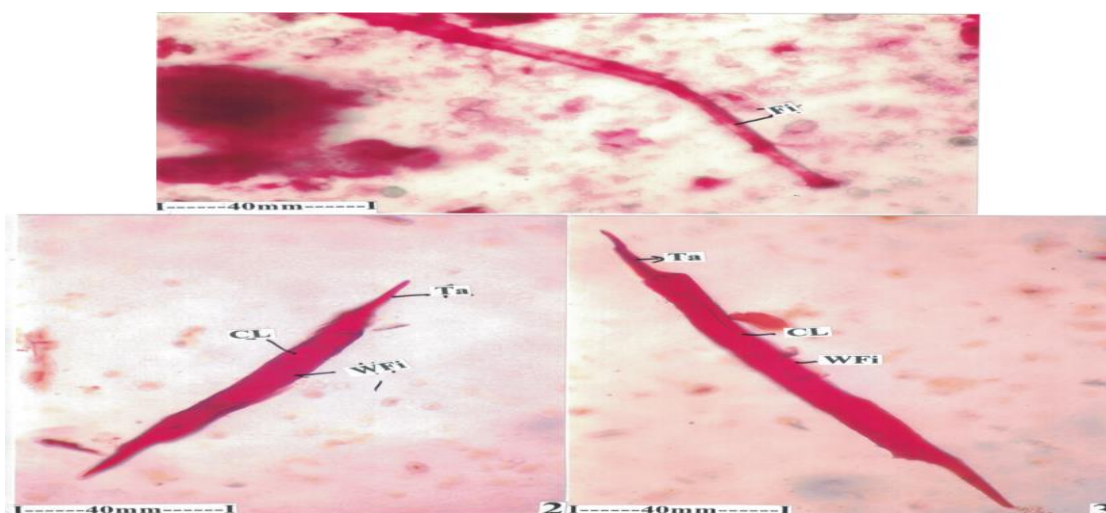


Figure: 5.10

1 & 2. The Powder consists of fibre long tapering ends with thin walled wide lumen.

3. There are also typical wide fibre, with thick middle part and suddenly narrow tapering ends of the fiber.

Fi – Fiber, CL-Cell lumen, Ta-Tail, WFi- Wide Fibre

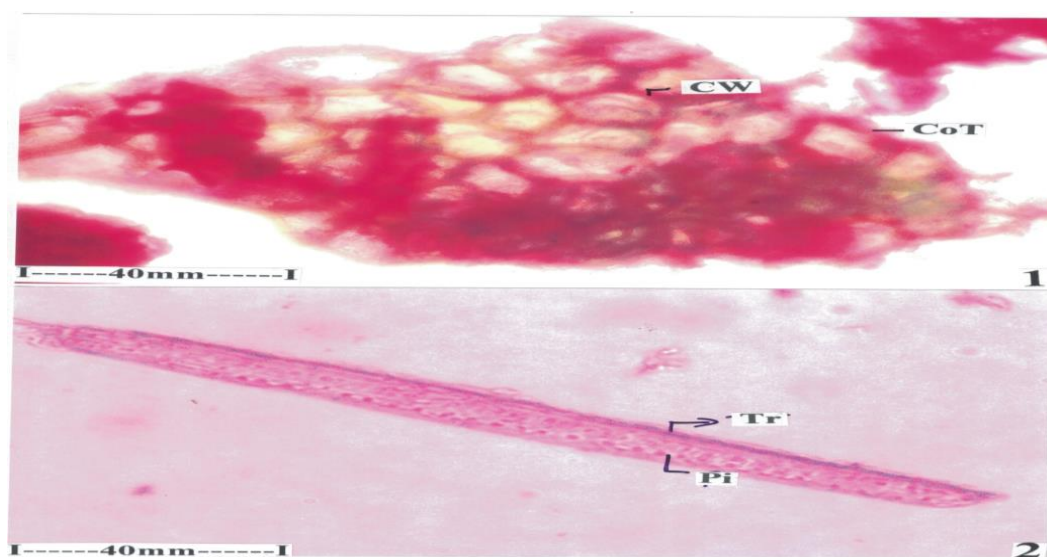


Figure 5.11

1. The small fragment are parenchymatous cells are also seen in the Powder. The cells are angular with thick walls

2. Tracheids are long cylindrical elements with numerous border pits with blunt end.

CW-Cell Wall, CoT- Cortical Tissue, Pi- Pits, Tr-Tracheid.

Physico-chemical Studies:

Various physico-chemical parameters were observed and recorded for the bark powder samples of *Terminalia paniculata* Roth. the results were depicted in Table 5.1

Table 5.1 Proximate values of *Terminalia paniculata* Roth. barks

Sl. No	Parameters		Obtained Values in %w/w
1	Moisture content	Loss on drying	9.5%
2	Values of Extractive	Extractives of Water soluble	5.90%
		Extractives of Alcohol soluble	9.04%
3	Ash Values	Total ash content	5.36%
		Content of Acid insoluble ash	0.75%
		Ash Water soluble	0.86%
		Ash Sulphated	1.43%

Preliminary qualitative phytochemical analysis:

The total yield of bark extracts obtained from *Terminalia paniculata* are mentioned in the Table 5.2. The phytochemical analysis of alcoholic and water extracts of *Terminalia paniculata* were carried out as per established procedure, to identify the various primary and secondary constituents such as sugars, proteins, amino acids, steroids, Saponin glycosides and Poly Phenolic compounds. The outcome of phytochemical results are mentioned in Table 5.3

Table 5.2 Values and physical characters of hydro ethanolic bark extract of *Terminalia paniculata* Roth.

Sl. No	Colour	Odour	Consistency	Yield (%w/w)
01	Reddish Brown	Characteristic	Crystalline	16.96 %

Table 5.3 Phytochemical analysis of hydro alcoholic bark extracts of *Terminalia paniculata*

Sl. no	Phytoconstituents	Hydro alcoholic bark extract
0	Sugars	Positive
1	Poly phenolic compounds	Positive
2	Flavonoids	Positive
3	Cardiac glycoside	Positive
4	Steroid	Positive
5	Triterpenoid	Positive
6	Coumarin	Positive

Analysis of poly phenolic compounds**Detection of Total Phenolic Content:**

The total content of phenol were determined using Foline-Ciocalteu method. Phenol content were analysed from the regression formula of the plot (standard)

($y = 0.0045x + 0.0042$, $R^2 = 0.9978$) and is mentioned as equivalents to gallic acid. The total content of phenol present in HBTP was found to be 89.42 ± 0.57 GAE mg/gm.

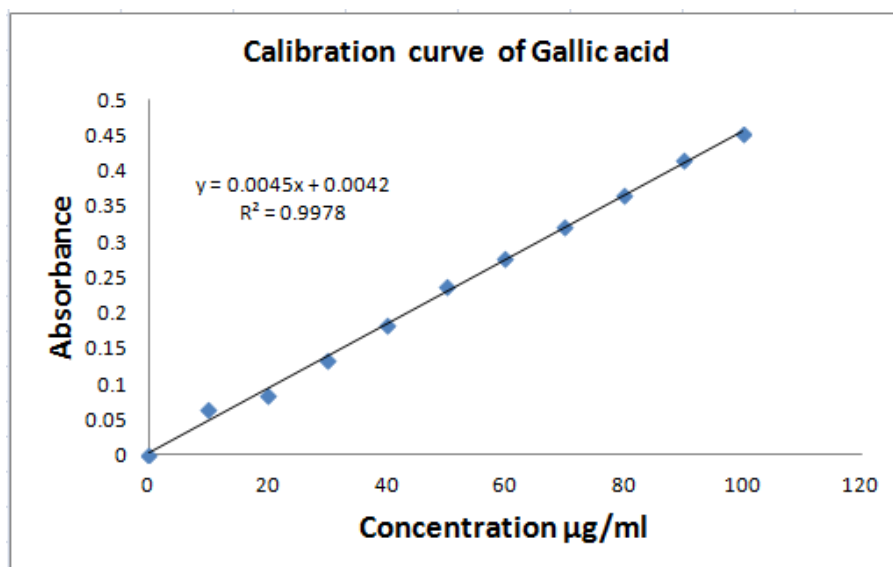


Figure 5.12 Standard calibration curve for total content of phenol for gallic acid (standard)

Analysis of Total Content of flavonoid:

The total content of flavonoid was determined using aluminum chloride method. Content of flavonoid was determined from the regression formula of the plot (standard) ($y = 0.0038x + 0.0013$, $R^2 = 0.9977$) and is mentioned as equivalents to quercetin. The total content of flavonoid present in HBTP was found to be 68.43 ± 1.53 QUE mg/gm.

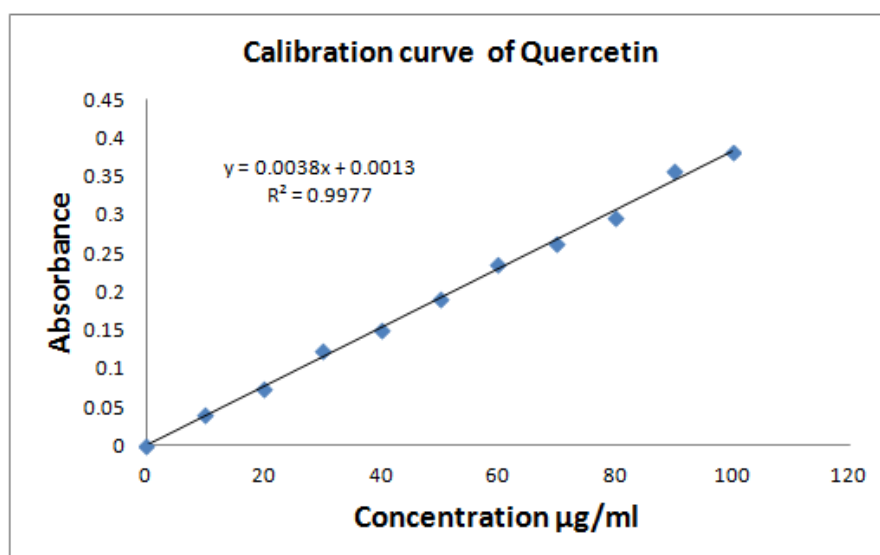


Figure 5.13 Calibration curve for total content of flavonoid for Quercetin (standard)

IN-VIVO ANTIULCER ACTIVITY:

A dose-response antiulcer activity was shown by 200 and 400 mg/kg of hydro ethanolic bark extract of *Termenalia paneculata* Roth. (HBTP) against ethanol-induced mucosal damage. The HBTP was given to various group of rats, orally, daily for 15 days and studies were carried out on 24 h fasted rats on 16th day. The result exhibited a dose- relevant mucoprotective activity in HBTP.

Effect of hydro ethanolic bark extracts of *Termenalia paneculata* Roth. in ethanol-administered mucoprotective activity.

Table 5.4 and Figure 5.14, 5.15 revealed dose dependent effect of HBTP. The mucoprotective effect of HBTP was found to increase with elevated doses. Ethanol treated animals showed rapid gastric ulcers. However, pre-treatment with HBTP decreased the severity and incidence of gastric ulcers in treated animals which was statistically significant. The ulcer index of inducing group was 6.24 ± 0.46 while that of test drug at (200mg/kg b.w) and (400mg/kg b.w) was $2.70 \pm 0.42^{**}$ ($p < 0.01$), $1.98 \pm 0.53^{***}$ ($p < 0.001$) and % inhibition was 53.61%, and 61.35% respectively. Standard drug sucralate ulcer index was found to be $1.10 \pm 0.63^{***}$ ($p < 0.001$) and % inhibition was 74.28%.

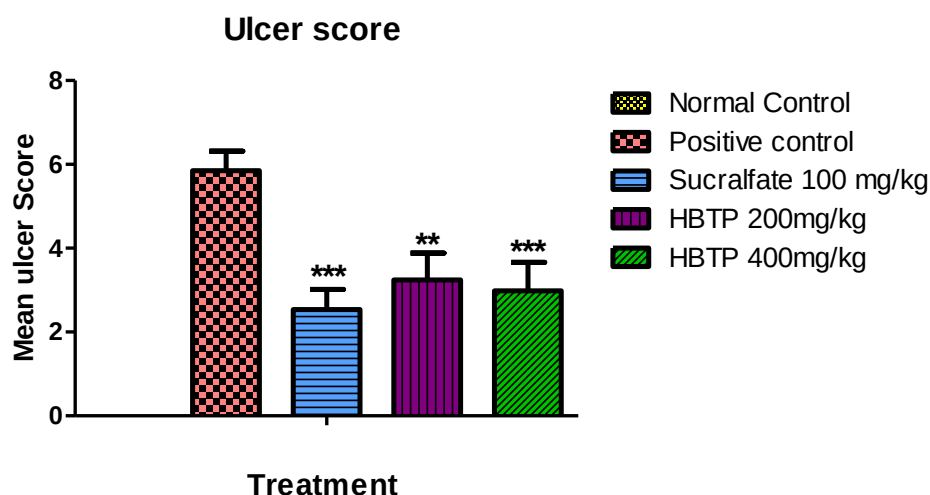
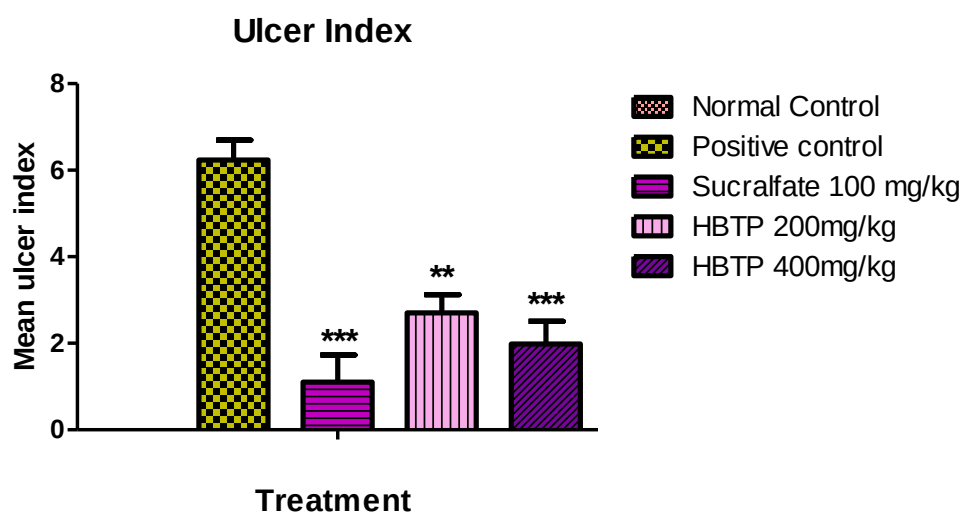
Morphological assessment of stomach:

The stomachs were got opened along with the bigger curvature and rinse with water to eradicate gastric content and blood clots and examined by a 10× magnifi lens to conclude the formation of ulcers. The numbers of ulcers were determined. Scoring of ulcer were mentioned as: Normal colored stomach (0), Red color (0.5), ulcer Spot (1), Hemorrhagic of streaks (1.5), Deep ulcerations (2), Perforations (3).

Table 5.4 Effect of hydro ethanolic bark extracts of *Termenalia paneculata* Roth. in ethanol induced ulcer index.

Sl. No	Groups	Score of Ulcer	Inhibition in ulcer score (%)	Index of Ulcer	Percentage protection index
1	Normal Control	--	--	0	100
2	Positive control 1ml/200gm (99%) ethanol orally	5.85±0.47	--	6.24±0.46	--
3	Standard drug Sucralfate 100 mg/kg, orally	2.54±0.48***	57.28	1.10±0.63***	74.28
4	HBTP Lower dose 200mg/kg + Ethanol	3.24 ±0.64**	46.27	2.70±0.42**	53.61
5	HBTP Higher dose 400mg/kg+ Ethanol	2.98±0.68***	54.89	1.98±0.53***	61.35

Expressed Values are as mean ± SEM (n = 6) and analyzed by ANOVA (one-way), by Tukey's post test. P values Compared with standard control and positive control, *p<0.05, **p<0.01, ***p<0.001 .

Figure 5.14 Effect of hydro ethanolic bark extract of *Termenalia paneculata* Roth. in ulcer score in ethanol induced ulcerFigure 5.15 Effect of hydro ethanolic bark extract of *Termenalia paneculata* Roth. in alcohol induced ulcer index

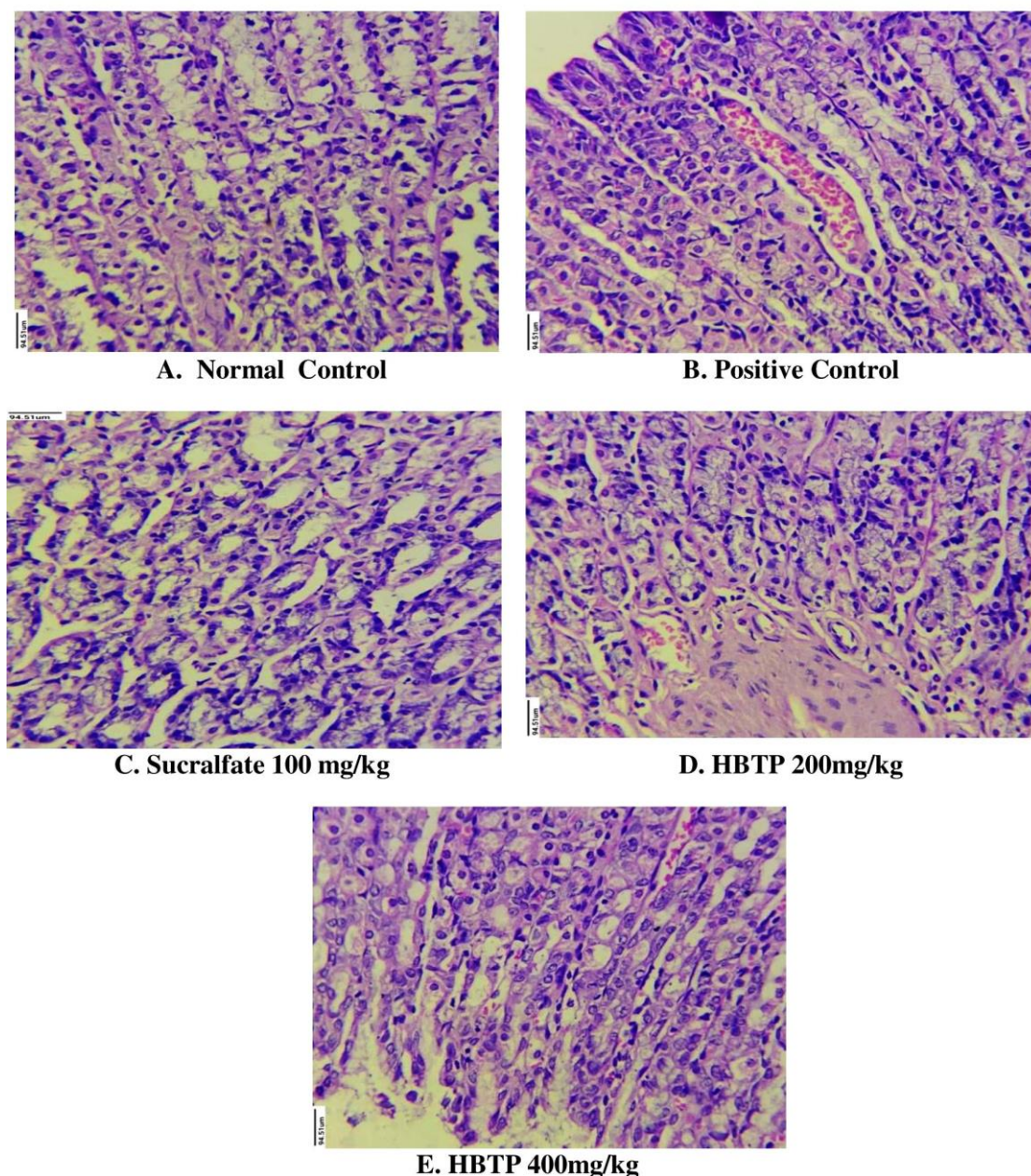


Figure 5.16 Histopathological observations of gastric mucosa

Effect of HBTP on gastric ulcer assessment:

The effect of HBTP on gastric volume and pH:

The analysis of pH for gastric juice, were employed to detect the efficiency of HBTP. The study shows that the normal control group has a pH of 4.08 ± 0.35 when compared with ulcer control group exhibiting a pH of 2.84 ± 0.25 . Sucralfate, HBTP 200 and 400mg/kg-administered animal groups showed pH values of $3.31 \pm 0.28^{**}$ and $3.53 \pm 0.44^{**}$ respectively.

The effect of HBTP on gastric acidity:

The free value of acidity of the ulcer control group was the higher 70.83 ± 2.38 mEq L⁻¹ amongst all the other treated groups. The acidity value for the normal control group was 31.22 ± 1.36 mEq L⁻¹, compared with $46.66 \pm 3.30^{***}$ mEq L⁻¹ in the standard drug (sucralfate) group. Further, the acidity values of the HBTP 200 and 400mg/kg groups were observed to be $48.74 \pm 1.32^{**}$ and $40.41 \pm 2.47^{***}$ respectively.

The total value of acidity of the ulcer control group was the higher 121.67 ± 7.03 mEq L⁻¹ amongst all the other treated groups. The acidity value for the normal control group was 40.66 ± 4.30 mEq L⁻¹, compared with $46.66 \pm 3.30^{***}$ mEq L⁻¹ in the standard drug (sucralfate) group. Subsequently, the acidity values of the HBTP 200 and 400mg/kg groups were observed to be $75.20 \pm 3.96^{**}$ and $56.04 \pm 3.08^{**}$. The results showed a reduced gastric volume and total acidity and raise in gastric pH with a significance of $^{**}p < 0.01$ and $P < 0.001$ for the sucralfate and HBTP treated groups.

The effect of HBTP on Mucin content:

In the control group of ulcer, the mucin content was 28.73 ± 2.05 mcg/g, whereas it was $53.36 \pm 1.53^{***}$ mcg/g in the case of the standard drug. The HBTP 200 and 400mg/kg groups exhibited mucus contents of $37.46 \pm 1.82^{**}$ and $48.07 \pm$

3.24***mcg/g. Thus, a rapid raise in mucous content was noted in HBTP 400mg/kg and standard drug sucralfate ($P < 0.001$).

Effect of hydro ethanolic bark extract of *Terminalia paniculata* Roth. On Histopathological profile:

The histology observations which were made on stomach of control and experimental rats are presented in Figure 5.16 (A-E). Figure 5.16A revealed the histological observations made on the gastric mucosa of normal control rats showing normal architecture. Figure 5.16B indicates stomach tissue of alcohol induced ulcer rats with rapid ulcer lesions on the gastric mucosa and infiltration of inflammatory cells. Figure 5.16C represent Sucralfate 100mg/kg treated rats shows less ulcer lesions. Figure 5.16D and E HBTP (200 and 400mg/kg) treated animals showed focal erosion and mild inflammatory cell infiltration indicating mucoprotective activity of the extract.

Discussion

Microscopic examination of bark of plant indicated the presence of periderm, Cortex and secondary Phloem, rhytidome collapsed phloem, non collapsed phloem, sieve elements, phloem rays (procumbent cell). Powder microscopy exhibited the presence of phloem fibre, parenchyma cells and tracheids.

Required amount of moisture content was found in air-dried materials of bark (9.5%). The extractive value is an essential physical-chemical parameter to determine the weight of crude extract and to detect the exhausted and adulterated plant materials[22]. Alcohol and water extractive values were found to be 9.04% and 5.90% respectively. The total value of ash for bark was 5.36%. values of acid-insoluble ash 1.75% indicates the presence of silicacious substances. Values of Water soluble ash are found to be 0.86%. The ash sulfated indicates the presence of inorganic impurities and ash sulfated was found to be 1.43%.

The phytochemical analysis of hydro alcoholic bark extract of *Terminalia paniculata* showed the presence of sugars, polyphenolic compounds, steroids, cardiac glycoside, Coumarins, triterpenoid. Triterpenoids are said to have cellprotective effect by increasing mucous production by increasing the PG, improving the blood flow of gastric, and increasing the secretion of gastric bicarbonate and mucous. In addition, terpenoid help as antioxidants, decrease the lipid peroxidation levels, and increase the of antioxidant enzymes activity in the gastric mucosa[23].

The overall analysis of total phenolic and flavonoid contents of HBTP was analysed by UV spectroscopic method. The total content of phenolic was found to be 89.42 ± 0.57 GAE mg/gm. The total content of flavonoid found to be 68.43 ± 1.53 QUE. These results showed that the *Terminalia paniculata* bark extract contain huge quantity of phenolic and flavonoid compounds.

In our experimental work, we noted a flattening in the stomach of mucosal wall of which indicates that the anti-ulcer effect of HBTP may be due to a reduced gastric motility. Rats treated with HBTP (200mg/kg and 400mg/kg) and those treated with sucralfate showed a good protection of their gastric mucosal content as observed by the reduction in the ulceration areas. The decreased submucosal fluids and the inflammatory reactions were also seen in these groups.

The experimental study reveals that the HBTP possesses significant anti-secretory, anti-ulcer and cellprotective properties in

rats. early-treatment with HBTP produced a dose-dependent decrease in the acidity and an increase in mucus contents. The mucoprotective activity of the extract was also identified by histology studies. Further these studies confirmed the effect of HBTP supplementation in preventing alcohol-induced hemorrhage and necrosis in the superficial layer of the gastric mucosa as indicated in Fig-5.16 (A-D). This dose also proves the most potential effects on ulcer index and the better protection. The cellprotective effect of the extracts may be due to their flavonoid content and by scavenging property. Certain agents, have the capacity to reduce gastric acid secretion, such as proton pump inhibitors (sucralfate) and histamine H₂-receptor antagonist, are relied to increase the healing process of the gastric lesions [24].

Conclusion

Our findings indicated that hydro alcoholic bark extract of *Terminalia paniculata* Roth. Will give protection to the gastric mucosa against alcohol induced injury. Such protective properties was shown to be dose related as observed by the decrease in the ulcer areas in the gastric wall as well as by the reduction of fluids and the leucocyte infiltration of layers of mucosal. Perhaps, dose of 400 mg/kg, HBTP protection could be due to balances between the acid-base production in the stomach itself and the mineral content of the plant. In further work, there is need to isolate and characterize the specific phytoconstituents of the plant extracts for ailment of peptic ulcer.

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