

EFFECT OF *PTEROCARPUS SANTALINOIDES* LEAF AQUEOUS EXTRACT ON STOMACH ETHANOL INDUCED ULCERATIVE ALBINO RATS

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Abstract: Background: *Pterocarpus santalinoides* leaves are valued for their therapeutic properties, nutritional relevance, and phytochemical richness, while peptic ulcer remains a common gastrointestinal disorder worldwide. Aim: This study investigated the effects of post-treatment with *P. santalinoides* leaf aqueous extract on ethanol-induced gastric ulcer in rats. Methodology: Thirty rats were acclimatized and divided into six groups: a normal control, a negative control (ethanol only), three treatment groups receiving 1500, 1000, and 700 mg/kg body weight of *P. santalinoides* extract, and a positive control treated with 20 mg/kg omeprazole. Following ethanol induction, extract treatments were administered orally for 14 days. Gastric tissues were harvested under light chloroform anesthesia, processed for oxidative stress parameters, and examined histologically using hematoxylin and eosin staining. Results: The findings showed that ethanol caused severe gastric mucosal injury, reflected in increased ulcer indices and lesion counts in the negative control group. Post-treatment with *P. santalinoides* extract produced a dose-dependent gastroprotective effect, with the high-dose group showing the greatest reduction in gastric damage and strongest antioxidant enhancement. Omeprazole treatment effectively reduced gastric acid secretion but primarily through acid suppression. These findings highlight the therapeutic potential of *P. santalinoides* as a natural gastroprotective agent against ethanol-induced gastric injury.

Keywords: *Pterocarpus santalinoides*, Ethanol, Stomach, Ulcer, Albino rats.

1. INTRODUCTION

Medicinal plants have long served as primary healing agents before the advent of modern pharmaceuticals (Srivastava, 2018). In Africa and other developing regions, a significant proportion of the population continues to rely on indigenous plants for healthcare (Mintah *et al.*, 2019). These plants are central to traditional medicine, with phytochemicals providing valuable leads for drug discovery (Shakya, 2016). Bioactive compounds such as flavonoids, polyphenols, and alkaloids exert antioxidant, anti-inflammatory, chemopreventive, and anticancer effects both in vitro and in vivo (Dar *et al.*, 2023; Asma *et al.*, 2022). Advances in phytochemistry, pharmacology, and biotechnology have further expanded research into plant-derived compounds and their therapeutic potential (Chaachouay *et al.*, 2024).

Pterocarpus santalinoides, a member of the Fabaceae family, is widely recognized in tropical Africa for its ethnomedicinal importance and ecological value (Ani, 2021; Ayéna *et al.*, 2021). It is known by various vernacular names; red sandalwood (English), nturukpa (Igbo), gbengbe (Yoruba), gunduru (Hausa), soro wisa (Akan, Ghana), fan dimi (Senegal), and ouokisse (French). The plant is native to Nigeria, Cameroon, Ghana, Sierra Leone, and Ivory Coast (Enemali *et al.*, 2019; Nair *et al.*, 2022).

Phytochemical studies revealed that the leaves contain alkaloids, tannins, flavonoids, saponins, and terpenoids (Chic *et al.*, 2014). Among Igbo-speaking communities in southeastern Nigeria, decoctions of the leaves and bark are used for stomach ache, diarrhea, and diabetes (Madubuike *et al.*, 2012). The Igede people in North Central Nigeria also consume leaf decoctions for infectious diseases (Chic *et al.*, 2014). More broadly, the plant has been applied in treating gastrointestinal disorders, microbial infections, inflammation, dysentery, fever, wounds, and pain (Keshavamurthy *et al.*, 2025).

Experimental studies have demonstrated additional therapeutic potential. Methanolic leaf extracts reduced protein loss in urine, suggesting restoration of glomerular integrity and renal protection (Ihedioha *et al.*, 2020). Furthermore, administration of the leaf, indicating a possible protective role against lipid-associated cardiovascular risks (Oyepata *et al.*).

Ethanol is a volatile, colorless, and flammable liquid with significant pharmacological and toxicological effects (Garg *et al.*, 2020). It's rapidly absorbed and distributed nature impacts multiple organ systems, especially the gastrointestinal tract, liver, and central nervous system (Salamah, 2025). Ethanol's physicochemical nature explains both its therapeutic utility and its cytotoxic potential, depending on concentration and context. (Brendan & Thomas, 2019).

In experimental research, ethanol is widely used to induce gastric mucosal injury due to its rapid and reproducible ulcerogenic effects (Yang *et al.*, 2017; Plaskova *et al.*, 2023). It extracts phospholipids from cell membranes, weakening mucosal integrity and facilitating hydrogen ion back-diffusion, which leads to epithelial damage, exfoliation, and necrosis (Ren *et al.*, 2020).

Oxidative stress is a major mechanism underlying ethanol-induced ulceration (Contreras-Zentella *et al.*, 2022). Ethanol also provokes microvascular disturbances such as reduced blood flow, vascular congestion, and increased capillary permeability, resulting in edema, hemorrhage, and ischemia (Mandyam *et al.*, 2026)

Gastric secretion involves hydrochloric acid (HCl), digestive enzymes, mucus, and intrinsic factor, regulated by neural, hormonal, and paracrine mechanisms. It occurs in three phases: cephalic, gastric, and intestinal (Hyder *et al.*, 2023; Furness, 2016).

Peptic ulcers are painful sores that develop in the stomach, esophagus, or duodenum, often slow to heal and recurrent (Ravisankar *et al.*, 2016). They result from an imbalance between aggressive factors (HCl, pepsin, *Helicobacter pylori*, NSAIDs) and defensive mechanisms (Liu *et al.*, 2016). Ulceration extends through the muscularis mucosae, driven by corrosive gastric acid and pepsin (Koikoibo, 2025). In this study, ethanol-induced ulcers occur by disrupting homeostasis, breaking the gastric mucosal barrier, and causing internal injuries.

Ethanol compromises mucosal defenses, provokes oxidative stress, impairs microvascular function, and stimulates inflammation. Collectively, these actions make ethanol a potent ulcerogenic agent and a widely accepted inducer in experimental peptic ulcer models.

2. MATERIALS AND METHODS

Collection of leaves

Fresh *Pterocarpus santalinoides* leaves were harvested from a farm in Aji, Enugu Ezike, Igbo-eze North Local Government Area, Enugu State, Nigeria. The freshly collected leaves were thoroughly washed with clean water and air-dried. Once dried, they were pulverized using a mechanical grinder to obtain fine leaf powder.

Extraction process

50 g of leaf powder was dissolved in 500 ml of water. The mixture was stirred, tightly covered, and stored overnight in a refrigerator. It was first sieved using muslin cloth, then filtered with Whatman No. 1 filter paper (125 mm). The filtrate was evaporated in an oven set at 50°C until a jelly-like concentrate was obtained. This concentrate was stored in a plastic container inside a refrigerator maintained at 4°C until required for experimental use.

Animal Study

Thirty (30) albino rats weighing between 160g-200g and of age 6 to 8 weeks were purchased and housed in the animal facility of Enugu State College of Medicine (ESUCOM) in well ventilated laboratory cages in groups of (6) with soft wood shavings as bedding. They were acclimatized for two weeks and allowed to have free access to food and clean water. The animals were maintained under laboratory conditions (temperature 24, with relative humidity of 60-70 percent, and a 12hour light-dark cycle). After the two weeks acclimatization, the rats were weighed before and after administration of substances.

The experiment was conducted according to the institutional ethics of animal use [ESUT/AREC/0225/37/2019030 190361MLS]

Study Design

Omeprazole reference studies have shown that administering 20 mg/kg body weight of omeprazole orally for 14 days can cure gastric ulcer (Akunna *et al.*, 2019). The LD50 of the aqueous extract of *Pterocarpus santalinoides* leaves is greater than 5000 mg/kg body weight in rats, indicating a high margin of safety (Ayéna *et al.*, 2022).

Ulcer induction

Out of thirty rats, twenty-five were induced with gastric ulcer immediately after acclimatization. Induction was achieved through oral administration of 5 ml per body weight of 95% ethanol.

Experimental groups

Group 1 was treated with baseline control and received pellet feed and water only, serving as the normal control. Group 2 (negative control) received pellet feed, water, and ethanol, representing untreated ulcer induction while group 3 (Positive control) Received pellet feed, water, ethanol, and 20 mg/kg body weight of omeprazole for 14 days, serving as the standard treatment group. Group 4, 5 and 6 were treated with pellet feed, water, ethanol, and (1500, 1000, 700) mg/kg body weight of *Pterocarpus santalinoides* extract for 14 days respectively.

Sample Collection and Analysis

At the twenty-ninth (29th) day, the animals were weighed then sacrificed under chloroform anesthesia. The stomach was identified and excised using sterile surgical blades. The organs were washed in normal saline (to remove excess blood) and a magnifying lens was used to count the number of ulcer score in each organ. After which the organs excised were divided into two, immediately fixed in buffer solution and in 10% neutral buffered formalin to avoid post-mortem changes.

Ulcerative Index

The harvested visceral organs were washed with normal saline and examined macroscopically using lens. Ulcer spots and streaks were recorded.

Oxidative Stress Parameter Analysis

The stomach tissues were broken into pieces in buffer solution 1:5 (wt/vol). Tissue 1g in 5ml of ice-cold phosphate buffer.

Histological Analysis

Tissue fixed in formalin were processed for histological studies and the sections stained using H and E staining method.

Statistical Analysis

The results of the test were analyzed using analysis of variance (ANOVA) and Tukey's post hoc test for multiple comparison with p value of ≤ 0.05 (95% confidence interval) bring considered as significant.

3. RESULTS

Ulcerative Index

Table 1 – Comparison of the Mean Number of Spots and Streaks across the Different Treatment Groups

Treatment groups	Spot	Streak
	Mean $\pm$ S.D	Mean $\pm$ S.D
High dose	3 $\pm$ 0.6	7.3 $\pm$ 2.3
Positive control	5 $\pm$ 1.7	8.7 $\pm$ 2.1
Medium dose	16 $\pm$ 2.1	13 $\pm$ 2.5
Negative control	16 $\pm$ 5.0	11 $\pm$ 5.7
Low dose	13 $\pm$ 2.8	16 $\pm$ 0.0
p-value	0.841	

Table 1 shows the comparison of the number of spots and streaks, which are indicators of gastric mucosal damage, across the different treatment groups. The high dose group recorded the lowest number of spots (3) and streaks (7), indicating the least ulcer severity and suggesting a strong protective effect of the extract at higher doses. The positive control group also

showed relatively fewer spots (5) and streaks (9), confirming the effectiveness of the standard anti-ulcer drug. In contrast, the medium dose and negative control groups had higher numbers of spots (16 each) and streaks (13 and 11, respectively), indicating more severe ulceration. The low dose group also showed considerable ulcer damage, with 13 spots and the highest number of streaks (16), suggesting limited protective effect at lower doses. However, the differences among the groups were not statistically significant ($p = 0.841$), indicating that although the high dose and positive groups showed fewer lesions, the variation across the treatment groups may not be sufficient to confirm a statistically significant effect.

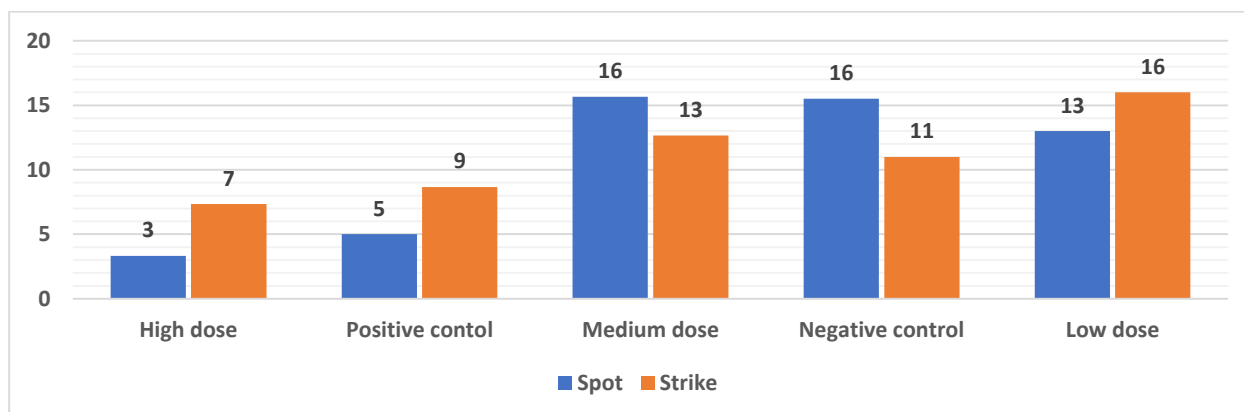


Fig 1 – Comparison of the number of spots and streaks across the different treatment groups

Table 2 – Ulcerative Index of the Different Treatment Groups

Treatment groups	Ulcerative index Mean $\pm$ S.D
High dose	8.2 $\pm$ 1.35
Positive control	10.8 $\pm$ 1.90
Medium dose	24.1 $\pm$ 3.40
Negative control	22.8 $\pm$ 8.7
Low dose	23.7 $\pm$ 2.83
p-value	0.009

Table 2 shows the ulcerative index of the different treatment groups, indicating varying degrees of gastric mucosal damage. The high dose group had the lowest ulcerative index (8.2), suggesting the strongest protective or healing effect against aspirin-induced ulceration. The positive control group also showed a relatively low ulcerative index (10.8), confirming the effectiveness of the standard anti-ulcer drug. In contrast, the medium dose (24.1), low dose (23.7), and negative control (22.8) groups had much higher ulcerative indices, indicating more severe ulceration and weaker protective effects. The similarity between the low and medium dose groups and the positive control suggests that lower doses of the extract provided limited protection. The difference across the groups was statistically significant ($p = 0.009$), indicating that the treatments, particularly the high dose of *Pterocarpus santalinoides* extract, had a significant effect in reducing ulcer severity. Overall, the findings suggest a dose-dependent anti-ulcer effect, with the high dose showing the most pronounced gastroprotective activity.

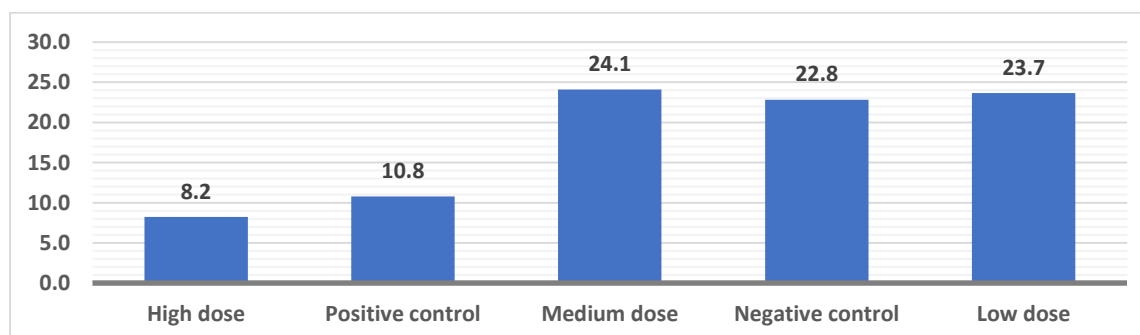


Fig 2 – Comparison of Ulcerative indexes of the treatment groups

Oxidative Stress Parameters

Table 3 – Comparison of the Oxidative Stress Levels Across all Treatment Groups

Treatment groups	MDA mg/dl	SOD U/L	CAT U/L	GSH mg/dl
	Mean $\pm$ SEM	Mean $\pm$ SEM	Mean $\pm$ SEM	Mean $\pm$ SEM
High dose	0.9 $\pm$ 0.13	9.7 $\pm$ 1.11	5.5 $\pm$ 0.21	10.2 $\pm$ 0.06
Medium dose	1.1 $\pm$ 0.03	8.9 $\pm$ 0.91	3.4 $\pm$ 0.58	9 $\pm$ 0.47
Low dose	1.3 $\pm$ 0.12	8.2 $\pm$ 0.61	2.9 $\pm$ 0.24	4.9 $\pm$ 0.54
Positive control	1.0 $\pm$ 0.03	10.9 $\pm$ 0.29	4.9 $\pm$ 0.05	8.9 $\pm$ 0.55
Normal control	1.2 $\pm$ 0.03	4.2 $\pm$ 0.07	1.4 $\pm$ 0.03	2.7 $\pm$ 0.28
Negative control	0.9 $\pm$ 0.15	9.8 $\pm$ 0.38	5.5 $\pm$ 0.70	10.2 $\pm$ 0.62
p-value	0.068	0.004	0.002	0.0001

Table 3 shows the comparison of oxidative stress markers across all treatment groups, including high, medium, and low doses of *Pterocarpus santalinoides*, the positive control, normal control, and negative control groups. The results indicate that antioxidant enzyme activities—superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH)—were generally higher in the high dose (SOD: 9.7 U/L, CAT: 5.5 U/L, GSH: 10.2 mg/dl) and positive control groups (SOD: 10.9 U/L, CAT: 4.9 U/L, GSH: 8.9 mg/dl) compared to the normal control group, which had the lowest antioxidant levels (SOD: 4.2 U/L, CAT: 1.4 U/L, GSH: 2.7 mg/dl), indicating increased oxidative stress in untreated ulcerated rats. The medium and low dose groups showed moderate improvements in antioxidant levels, suggesting a dose-dependent effect. Malondialdehyde (MDA) levels, a marker of lipid peroxidation, showed no statistically significant difference across groups ($p = 0.068$), although slightly lower values were observed in the high dose, positive control, and negative control groups. However, there were statistically significant differences in SOD ($p = 0.004$), CAT ($p = 0.002$), and GSH ($p = 0.0001$), indicating that the treatments, particularly the high dose of the extract and the standard drug, significantly improved antioxidant defenses. Overall, these findings suggest that *Pterocarpus santalinoides* leaf extract exerts protective effects against aspirin-induced oxidative stress by enhancing antioxidant enzyme activity.

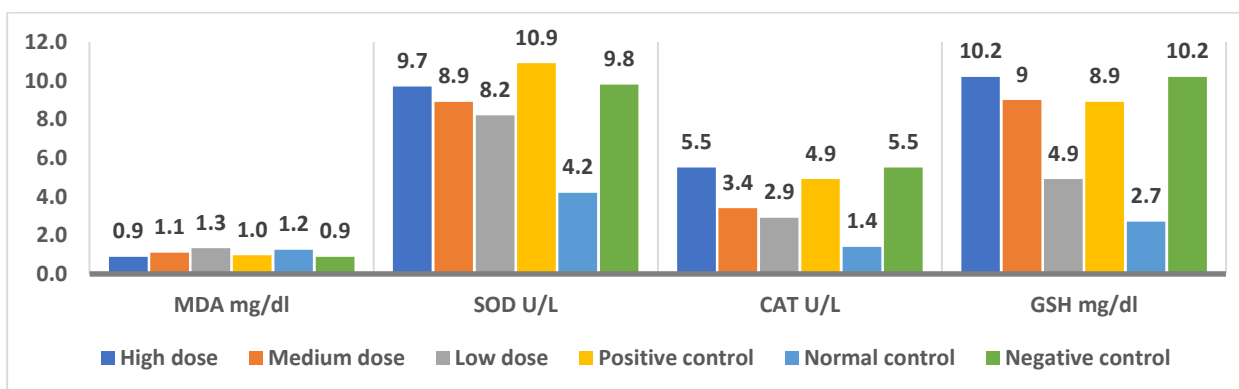


Fig 3 – Oxidative index levels across treatment groups

Histomorphological Findings

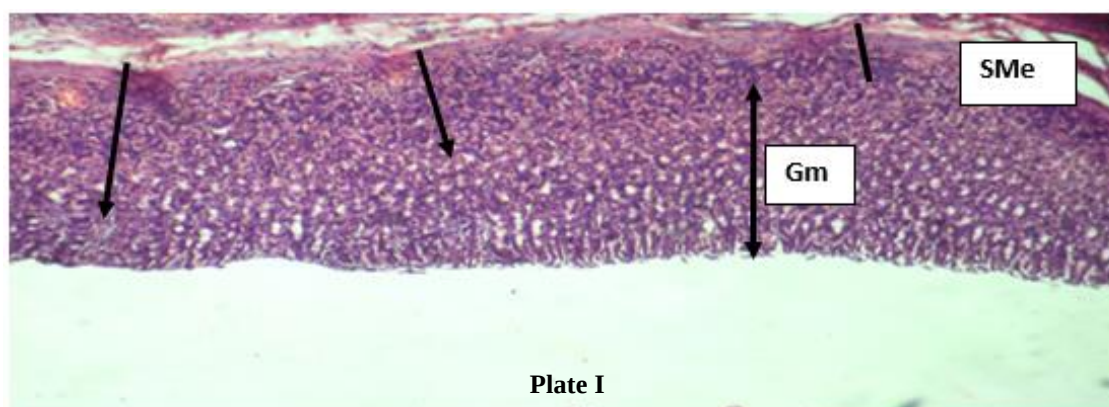


PLATE I - Light photomicrograph of Stomach tissue from rat post-treated with 20mg Omeprazole (positive control-group1) after induction of gastric injury with ethanol showing moderate gastro-protection, the gastric mucosa (Gm) is intact. However, erosion of some portions of the Submucosa (SMe) and marked cellular infiltration (arrows) are observed. Stain: Haematoxylin and eosin. Magnification: X100.

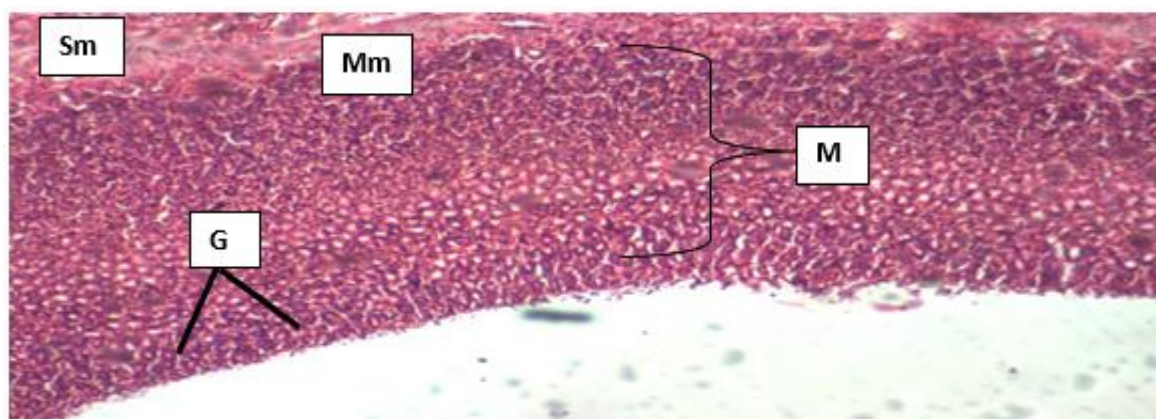


PLATE II

PLATE II - Light photomicrograph of representative stomach section of rats in the normal control (group 2) showing normal histological features of the mucosa (M) bearing the gastric gland (G), muscularis mucosa (Mm) and submucosa (Sm). Stain: Haematoxylin and eosin. Magnification: X100.

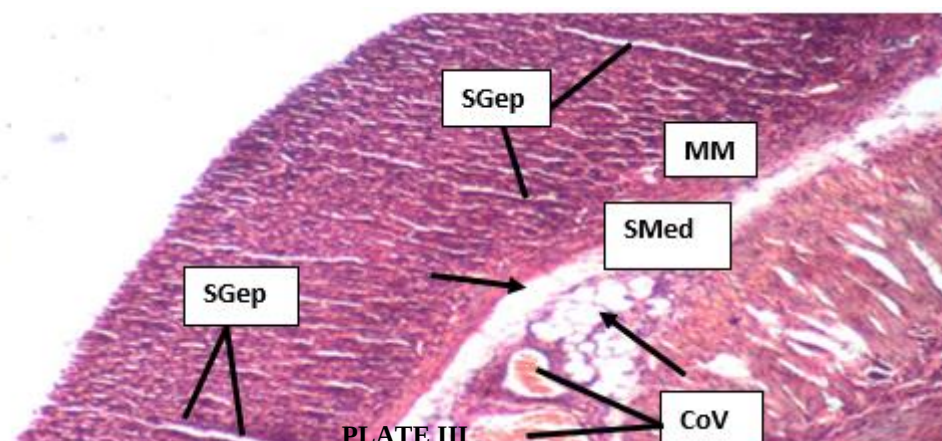


PLATE III

PLATE III: Light photomicrograph of Stomach section from negative control rat (Group 3) treated with 5ml of 95% body weight of ethanol only showing obvious histoarchitectural abnormalities of the gastric tissue. There is shedding of some areas of the gastric epithelial cells (SGep), submucosal edema (SMed), submucosal erosion (arrows) and congested vessels (Cov) (Stain: H&E; Mag: x100).

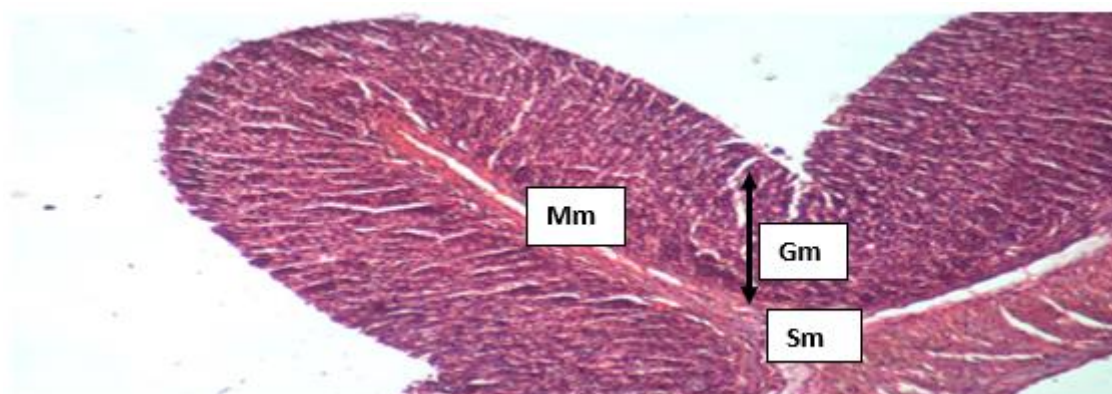


PLATE IV

PLATE IV: Light photomicrograph of Stomach section from rat post-treated with high dose 1500mg/kg body weight of *Pterocarpus santalinoides* (Group 4) following ethanol induced gastric toxicity showing good preservation of the gastric tissues. The gastric mucosa (Gm), submucosa (Sm) and muscularis mucosa (Mm) appear normal (Stain: H&E; Mag: x100).

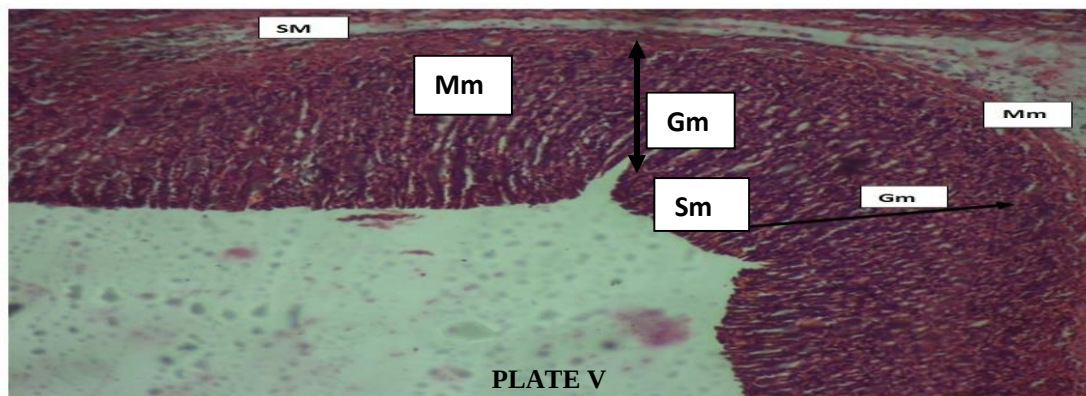


PLATE V: Light photomicrograph of Stomach section from rat post-treated with medium dose 1000mg/kg body weight of *Pterocarpus santalinoides* (Group 5) following ethanol induced gastric toxicity showing good preservation of the gastric tissues. The gastric mucosa (Gm), sub mucosa (Sm) and muscularis mucosa (Mm) appear normal. (Stain: H&E; Mag: x100).



PLATE VI: Light photomicrograph of Stomach section from rat post-treated with low dose 700mg/kg body weight of *Pterocarpus santalinoides* leaf extract (Group 6) following ethanol induced gastric toxicity showing fair preservation of the gastric tissues. The gastric mucosa (Gm) and muscularis mucosa (Mm) appear normal. However, submucosal erosion (SMer) and mild inflammatory cellular infiltration (arrows) are observed. (Stain: H&E; Mag: x100).

4. DISCUSSION

Ethanol administration is well known to induce acute gastric mucosal injury through mechanisms involving oxidative stress, mucosal necrosis, vascular damage, and inflammatory responses (Al Asmari, *et al.*, 2016). In this study, the negative control group exposed to ethanol alone exhibited pronounced gastric lesions, as reflected by elevated ulcerative indices and increased numbers of mucosal spots and streaks, confirming successful induction of gastric injury. Post-treatment with *Pterocarpus santalinoides* leaf extract resulted in varying degrees of gastroprotection. The high-dose group (1500 mg/kg) demonstrated the lowest ulcerative index (8.2), indicating a marked reduction in ulcer severity. This protective effect was comparable to, and slightly better than, that observed in the omeprazole-treated group (ulcerative index: 10.8). In contrast, the medium- and low-dose extract groups exhibited higher ulcerative indices, suggesting weaker protection at lower doses. The statistically significant difference observed among treatment groups ($p = 0.009$) supports a dose-dependent anti-ulcer effect of the extract. Although lesion counts (spots and streaks) did not show statistical significance ($p = 0.841$), the numerical reduction in the high-dose and drug control groups remains biologically relevant. The lack of statistical significance may be attributed to inter-animal variability or sample size limitations, rather than absence of therapeutic effect. The observed gastroprotective activity of *Pterocarpus santalinoides* aligns with previous studies reporting anti-ulcer

properties of the plant, which have been attributed to its rich phytochemical composition, including flavonoids, tannins, phenolics, and saponins (Chic, *et al.*, 2014). These compounds are known to enhance mucosal defense, scavenge free radicals, and stabilize gastric epithelial integrity.

The negative control group exhibited significantly reduced levels of antioxidant enzymes (SOD, CAT, and GSH), indicating severe oxidative stress and compromised mucosal defense. Conversely, treatment with *Pterocarpus santalinoides* extract significantly elevated antioxidant parameters, particularly at high doses. The high-dose extract group showed notable increases in SOD (9.7 U/L), CAT (5.5 U/L), and GSH (10.2 mg/dl), comparable to values observed in the omeprazole-treated group. The statistically significant differences recorded for SOD ($p = 0.004$), CAT ($p = 0.002$), and GSH ($p = 0.0001$) confirm that the extract effectively enhanced antioxidant defenses. These findings suggest that the gastroprotective effect of the extract is partly mediated through attenuation of oxidative stress. Malondialdehyde (MDA), a marker of lipid peroxidation, showed no statistically significant variation ($p = 0.068$), although lower levels were observed in the high-dose and drug-treated groups. This trend nonetheless supports reduced membrane lipid damage following extract administration. The antioxidant activity of *Pterocarpus santalinoides* has been widely documented and is linked to its high phenolic and flavonoid content. These compounds neutralize ROS, inhibit lipid peroxidation, and preserve cellular integrity, thereby mitigating gastric mucosal injury (Zhang, *et al.*, 2020).

The normal control group displayed intact gastric architecture, characterized by well-organized mucosa, preserved gastric glands, intact muscularis mucosa, and normal submucosa. This confirms absence of pathological alterations under physiological conditions. In contrast, the negative control group exhibited severe histoarchitectural disruption, including shedding of gastric epithelial cells, submucosal edema, vascular congestion, and submucosal erosion. These lesions are typical of ethanol-induced gastric damage and reflect epithelial necrosis, microvascular injury, and inflammatory responses. Ethanol is known to compromise mucosal barrier function, increase vascular permeability, and trigger inflammatory infiltration, culminating in tissue destruction (Mak, *et al.*, 2025). The positive group demonstrated moderate gastroprotection, with preservation of mucosal integrity but evidence of submucosal erosion and marked inflammatory cellular infiltration. While omeprazole effectively reduces gastric acid secretion, it may not completely prevent ethanol's direct cytotoxic effects, explaining the residual inflammatory changes observed. The high-dose group (1500 mg/kg) showed near-normal gastric morphology, with intact mucosa, preserved submucosa, and normal muscularis mucosa. The absence of significant erosion, edema, or inflammatory infiltration indicates strong cytoprotective and mucosal healing effects. The medium-dose group (1000 mg/kg) also demonstrated good preservation of gastric layers, suggesting substantial protective activity, though slightly less pronounced than the high-dose group. The low-dose group (700 mg/kg) exhibited fair mucosal preservation but mild erosion and inflammation indicated partial protection consistent with higher ulcer scores. This indicates partial protection, consistent with the higher ulcerative indices recorded at lower doses.

5. CONCLUSION

Pterocarpus santalinoides leaf extract showed strong gastroprotective and antioxidant effects in albino rats with ethanol-induced gastric injury. At higher doses, it significantly reduced ulcer severity, boosted antioxidant defenses (SOD, CAT, GSH), and maintained gastric tissue integrity. Its effects were similar to omeprazole, likely through reducing oxidative stress, strengthening the gastric barrier, and lowering inflammation. These findings highlight its potential as a natural treatment for gastric ulcers.

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