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Oxidative Stress and Renal Dysfunction in Lincomycin-Induced Nephrotoxicity: Evaluating the Therapeutic Potential of Activated Charcoal

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Abstract

Background: The kidneys play a vital role in homeostasis and metabolic waste elimination, but they are highly susceptible to toxic insults due to their role in drug metabolism. Lincomycin, a lincosamide antibiotic, has been implicated in nephrotoxicity through oxidative stress-mediated mechanisms, leading to renal dysfunction. Activated charcoal, a widely used adsorbent, has shown potential in mitigating renal damage by adsorbing toxins and modulating oxidative stress. However, its efficacy in lincomycin-induced nephrotoxicity remains poorly understood. ***Aim:*** This study investigates the protective potential of activated charcoal against lincomycin-induced nephrotoxicity by assessing oxidative stress markers, renal function indices, and histopathological changes. ***Methodology:*** Twenty-five (25) Wistar rats were divided into five groups (n=5). Group I (Control) received normal saline, while Group II received lincomycin (200 mg/kg). Groups III, IV, and V were co-administered lincomycin with varying percentages of activated charcoal (25%, 50%, and 75%). Kidney function markers (creatinine, urea), oxidative stress indices (Superoxide Dismutase [SOD], Malondialdehyde [MDA]), and histopathological changes were evaluated. ***Results:*** Lincomycin administration significantly reduced creatinine (0.59±0.07 mg/dl) and urea (19.85±2.11 mg/dl) compared to controls (0.85±0.04 mg/dl, 25.78±1.19 mg/dl; $P<0.05$). Oxidative stress was evident in the lincomycin group, with a decrease in SOD (14.25±1.81 U/mg protein) and an increase in MDA. Activated charcoal co-administration mitigated these effects, improving kidney function and oxidative stress parameters. ***Conclusion:*** Activated charcoal offers protective effects against lincomycin-induced nephrotoxicity by reducing oxidative stress and preserving renal function. Its potential as an adjunct therapy in mitigating antibiotic-induced kidney damage warrants further investigation.

Keywords: nephrotoxicity, lincomycin, activated charcoal, oxidative stress, renal dysfunction, antioxidant therapy

1. Introduction

The kidneys are vital organs that play a critical role in human physiology by maintaining fluid and electrolyte homeostasis, regulating blood pressure, supporting erythropoiesis, and ensuring efficient waste elimination. They are structurally adapted for filtration and selective reabsorption, which are crucial processes in metabolic waste removal and acid-base balance (Lote, 2012). Despite their anatomical and physiological significance, the kidneys are highly susceptible to toxic insults due to their role in drug metabolism and excretion. Nephrotoxicity, a condition characterized by kidney dysfunction caused by exposure to harmful substances, has emerged as a major concern in pharmacotherapy, particularly in the context of antibiotics and other nephrotoxic agents (Wang

et al., 2021).

Among nephrotoxic antibiotics, lincomycin, a lincosamide antibiotic primarily used for treating serious bacterial infections, has been associated with adverse renal effects (Liu *et al*., 2011). While lincomycin is often reserved for cases where penicillin is contraindicated, its potential to induce nephrotoxicity through oxidative stress and inflammatory pathways is an area of growing concern (Zhang *et al*., 2022). Lincomycin-induced nephrotoxicity is thought to result from mitochondrial dysfunction, increased reactive oxygen species (ROS) production, and subsequent oxidative damage to renal cells. Long-term exposure to lincomycin may exacerbate kidney injury by impairing antioxidant defense mechanisms, leading to structural and functional deterioration of the renal parenchyma (Chen *et al*., 2023).

Oxidative stress is a well-recognized pathological mechanism underlying kidney injury, contributing to renal inflammation, apoptosis, and fibrosis. Excessive Reactive Oxygen Species (ROS) production can overwhelm the antioxidant capacity of renal cells, resulting in lipid peroxidation, protein oxidation, and DNA damage (Gong *et al*., 2021). The role of oxidative stress in nephrotoxicity has been widely studied, with recent evidence highlighting the importance of therapeutic strategies that target ROS neutralization to mitigate renal damage (Huang *et al*., 2022). This has led to increased interest in natural and synthetic antioxidants as potential nephroprotective agents.

Activated charcoal, a well-known adsorbent with a high surface area and strong binding capacity, has been explored for its detoxification properties in various clinical settings (Zhao *et al*., 2023). Traditionally used for treating poisonings and overdoses, activated charcoal functions by binding to toxins in the gastrointestinal tract, preventing their systemic absorption and facilitating their excretion (WHO, 2009). Recent studies suggest that activated charcoal may also have renoprotective effects by adsorbing uremic toxins and reducing oxidative stress markers in renal dysfunction (Sharma *et al*., 2020). Its ability to modulate the gut-kidney axis and reduce systemic inflammation further underscores its potential as an adjunct therapy in kidney-related disorders (Kim *et al*., 2021).

Despite the known nephrotoxic effects of lincomycin and the emerging interest in activated charcoal as a potential nephroprotective agent, there is limited research evaluating its efficacy in mitigating lincomycin-induced renal injury. Current studies have primarily focused on the role of antioxidants and adsorbents in general nephrotoxicity models, leaving a gap in understanding their specific impact on lincomycin-induced oxidative stress and renal dysfunction. Additionally, while oxidative stress has been implicated in various nephropathies, its precise contribution to lincomycin-induced nephrotoxicity and the extent to which activated charcoal can counteract these effects remain unclear (Zhao *et al*., 2023).

This study aims to bridge this gap by evaluating the therapeutic potential of activated charcoal in lincomycin-induced nephrotoxicity. By investigating key oxidative stress markers, renal function indices, and histopathological changes, this research seeks to provide insights into the protective mechanisms of activated charcoal against antibiotic-induced renal damage. The findings from this study will not only contribute to the existing body of knowledge on nephrotoxicity mitigation but also offer a potential therapeutic strategy for preserving kidney function in patients undergoing lincomycin therapy. Given the increasing prevalence of antibiotic-associated kidney injury, identifying effective interventions to prevent or reverse renal dysfunction is of paramount clinical significance.

2. Materials & Methodology

2.1 Experimental Animals

Twenty-five (25) healthy adult male Wistar rats, weighing between 135.4 and 159.9 g, were procured for the study. The animals were housed in the Animal House of the College of Health Sciences, Benue State University, Makurdi. They were acclimatized for fourteen (14) days before the commencement of the experiment. The rats were randomly assigned into five (5) groups, with five (5) rats per group, and housed in plastic cages under standard laboratory conditions at a temperature of 28-31°C. They were provided with standard vital rat feed and water *ad libitum* throughout the experimental period.

2.2 Experimental Drug

Lincomycin capsules were obtained from Mernax Pharmacy, opposite the College of Health Sciences, Benue State University, Makurdi. The drug was administered orally via gavage after dissolving in distilled water.

2.3 Preparation of Activated Charcoal

Activated charcoal was derived from wood charcoal collected from a domestic kitchen in Otukpa, Ogbadibo Local Government Area, Benue State. The charcoal was thoroughly rinsed with water to remove debris, dried, and ground into fine powder. To activate the charcoal, 1.6 g of the powdered charcoal was soaked in a solution of 64 g of calcium chloride dissolved in water for 24 hours. Afterward, the mixture was filtered, and the charcoal

was subjected to thermal activation by heating over a gas flame until fully dried. The activated charcoal was then sieved to obtain fine particles, which were stored in airtight containers for use in the experiment.

2.4 Housing and Feeding Conditions

The rats were housed in five (5) plastic cages, ensuring adequate space for movement. Vital animal feed was purchased from a feed store in Wurukum, Makurdi, and stored at an optimal temperature in the animal house to maintain freshness.

2.5 Experimental Design

The study was conducted over 14 days following the acclimatization period. The 25 Wistar rats were divided into five (5) experimental groups, each consisting of five (5) rats, and were treated as follows:

- **Group I (Control Group):** Received 5 ml/kg body weight of normal saline orally at 12-hour intervals for 14 days.
- **Group II:** Received 200 mg/kg body weight of Lincomycin orally at 12-hour intervals for 14 days.
- **Group III:** Received a diet consisting of 75% standard feed mixed with 25% activated charcoal, along with water and 200 mg/kg of Lincomycin, administered at 12-hour intervals for 14 days.
- **Group IV:** Received a diet consisting of 50% standard feed mixed with 50% activated charcoal, along with water and 200 mg/kg of Lincomycin, administered at 12-hour intervals for 14 days.
- **Group V:** Received a diet consisting of 25% standard feed mixed with 75% activated charcoal, along with water and 200 mg/kg of Lincomycin, administered at 12-hour intervals for 14 days.

2.6 Animal Sacrifice and Sample Collection

At the end of the 14-day experimental period, the animals were humanely sacrificed using chloroform anesthesia. Blood samples were collected via cardiac puncture using sterile disposable syringes and stored in EDTA bottles for biochemical analysis. The kidneys were excised and fixed in 10% formal saline for histological examination and tissue processing.

2.7 Biochemical Analyses

2.7.1 Creatinine Determination

Serum creatinine levels were measured using the modified Jaffe method with the Quimica Clinica Applicada (QCA) creatinine test kit. In this method, creatinine in an alkaline solution reacts with picrate to form a colored complex. The rate of absorbance increase at 546 nm was measured and was directly proportional to the creatinine concentration, expressed in mg/dL.

2.7.2 Urea Determination

Serum urea concentration was estimated using the modified Searcy method with the Quimica Clinica Applicada (QCA) enzymatic urea test kit (Diamond Diagnostic, Hanover, Germany). The urea concentration was expressed in mg/dL.

2.7.3 Estimation of Lipid Peroxidation (Malondialdehyde, MDA)

Lipid peroxidation in renal tissues was assessed colorimetrically using the thiobarbituric acid reactive substances (TBARS) method of Buege and Aust (1978). A principal component of TBARS, malondialdehyde (MDA), is a marker of lipid peroxidation. Briefly, 0.1 mL of tissue homogenate in Tris-HCl buffer (pH 7.5) was mixed with 2 mL of a 1:1:1 solution of thiobarbituric acid (0.37%), trichloroacetic acid (15%), and hydrochloric acid (0.25 N). The mixture was incubated in a water bath at 95°C for 15 minutes, cooled, and centrifuged. The absorbance of the supernatant was measured at 535 nm against a blank. MDA concentration was calculated using its molar absorptivity (1.56 x 105 M-1cm-1) and expressed as nmol/mg protein.

2.7.4 Superoxide Dismutase (SOD) Activity Assay

Superoxide dismutase (SOD) activity was measured following the method of Winterbourn (1975), as described by Rukmini *et al*. (2004). This assay is based on the ability of SOD to inhibit the reduction of nitro-blue tetrazolium (NBT). The reaction mixture contained 2.7 mL of 0.067M phosphate buffer (pH 7.8), 0.05 mL of 0.12mM riboflavin, 0.1 mL of 1.5mM NBT, 0.05 mL of 0.01M methionine, and 0.1 mL of enzyme extract. The tubes were uniformly illuminated in an aluminum foil-lined box with a 15W fluorescent lamp for 10 minutes. A control reaction lacking the enzyme extract was also included. The absorbance was measured at 560 nm. One unit of SOD activity was defined as the amount of enzyme required to inhibit NBT reduction by 50%. Results were expressed as units/mg protein.

2.7.5 Histological Tissue Processing

Kidney tissues were embedded in molten paraffin wax and allowed to solidify in metallic tissue molds. The

blocks were then cooled at 5°C for 15 minutes, removed from the molds, and trimmed. Serial sections (3 µm thick) were obtained using a rotary microtome and floated in a water bath at 55°C. The sections were mounted onto clean frosted-end slides, placed on a hot plate for 40 minutes for proper adhesion, and then deparaffinized, hydrated, air-dried, and stored for staining.

2.7.6 Haematoxylin and Eosin (H&E) Staining

1) Sections were dewaxed in xylene (3 changes, 5 min each).
2) Rehydration was performed through descending ethanol concentrations (absolute, 95%, 80%, and 70%).
3) Staining was carried out using Harris hematoxylin (5 min).
4) Sections were rinsed in running tap water to remove excess stain.
5) Differentiation was performed in 1% acid alcohol (3 sec).
6) Sections were blued in running tap water (10 min).
7) Counterstaining with 1% eosin was done (1 min).
8) Dehydration was achieved through ascending ethanol concentrations (70%, 80%, 95%, and absolute).
9) Sections were cleared in xylene, air-dried, and mounted with dibutyl phthalate polystyrene xylene (DPX).

Slides were examined under a light microscope, and photomicrographs were captured.

2.8 Statistical Analysis

Data were analyzed using the Statistical Package for the Social Sciences (SPSS) version 23. Mean values were compared using one-way analysis of variance (ANOVA), and intergroup comparisons were performed using the least significant difference (LSD) post-hoc test. A p-value of <0.05 was considered statistically significant.

2.9 Ethical Considerations

This study was conducted in compliance with the ethical guidelines of the Ethical Committee of the College of Health Sciences, Benue State University, Makurdi. Approval was obtained before the commencement of the research.

3. Results

3.1 Body Weight Changes

The results of the body weight changes across the different groups show a significant variation in weight differences as presented in Figure 1. Group I (Control) had a mean weight increase of 22.10±10.81 g, while Group II (Negative Control: Lincomycin) showed a significantly higher weight increase of 38.26±17.98 g (P=.03 compared to Control), indicating a positive effect of Lincomycin on body weight gain. Group III, with a 75% feed and 25% activated charcoal (AC) mix, showed a moderate weight gain of 23.62±13.86 g, which was not significantly different from the control. Group IV, with a 50% feed and 50% AC mix, had a smaller weight increase of 14.82±6.51 g (P=.02 compared to the Negative Control), suggesting a potential inhibitory effect of the higher AC content. Group V (25% feed + 75% AC) showed a weight decrease of -8.56±6.74 g ($P<0.05$ compared to both Control and Negative Control), highlighting a significant negative impact of higher AC content on body weight.

The results suggest that while Lincomycin enhances body weight, the inclusion of activated charcoal, particularly in high amounts, negatively affects body weight gain.

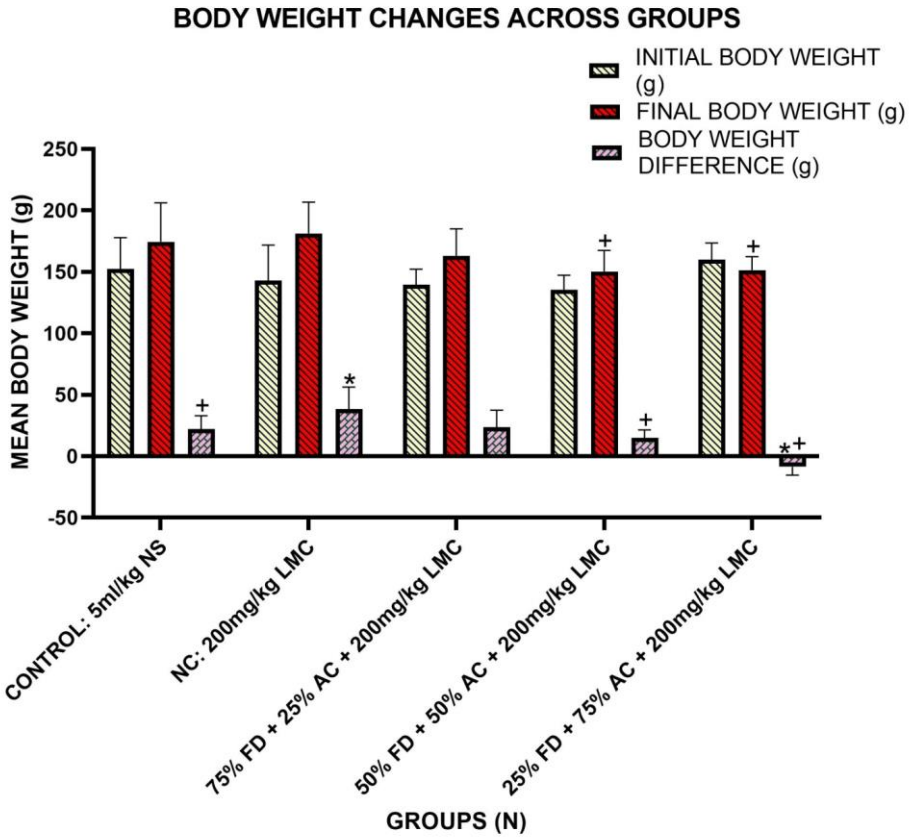

Figure 1. Simple Bar Chart Showing the Mean Body Weight Changes across Groups

N = 5; NC = Negative Control; LMC = Lincomycin; AC = Activated Charcoal; FD = Feed; *P<0.05 Compared to the Control Group; +P<0.05 Compared to the Negative Control Group.

3.2 Kidney Weight

The results presented in Figure 2 show the mean kidney weight and kidney/body weight ratio across groups. The kidney weight was consistent across all groups, with a mean of 1.70±1.30 g. However, the kidney/body weight ratio showed notable variation, particularly in Group V (25% Feed + 75% Activated Charcoal + 200 mg/kg Lincomycin), which had a significantly lower ratio of -0.84±1.49, compared to both the control and negative control groups (P=.01).

This suggests that the combination of activated charcoal and Lincomycin at this specific dosage and feed ratio might have a pronounced impact on kidney function or morphology, potentially altering the kidney's relative mass in relation to body weight. No significant differences were observed in kidney weight among all groups, indicating that the variations in the kidney/body weight ratio may reflect physiological changes induced by the treatments, particularly in Group V.

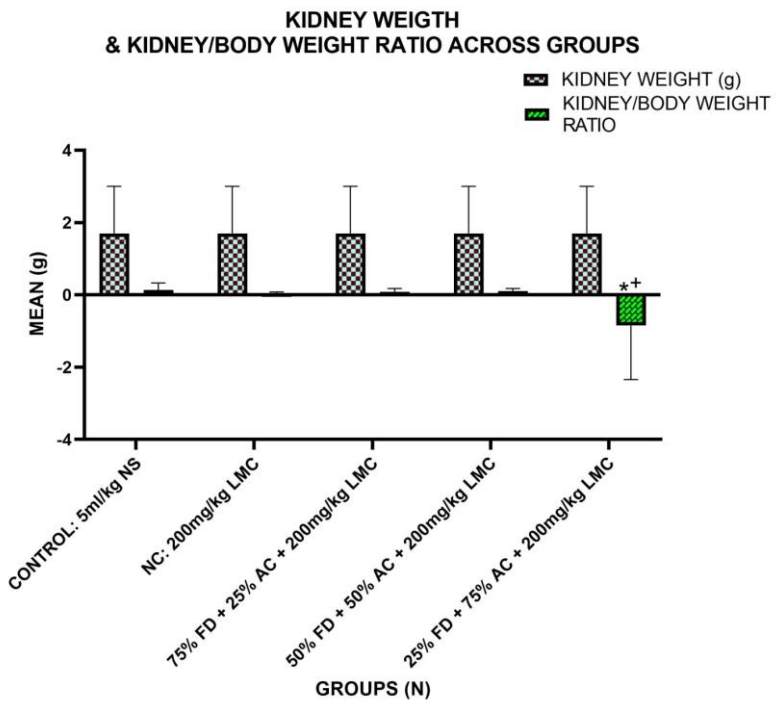

Figure 2. Simple Bar Chart Showing the Mean Body Weight & Body/Kidney Weight Ratio across Groups

N = 5; NC = Negative Control; LMC = Lincomycin; AC = Activated Charcoal; FD = Feed; *P<0.05 Compared to the Control Group; +P<0.05 Compared to the Negative Control Group.

3.3 Kidney Function Parameters

The results presented in Figure 3 show the mean kidney function parameters (creatinine and urea levels) across groups. Group I (Control) showed a creatinine level of 0.85±0.04 mg/dl and urea level of 25.78±1.19 mg/dl. Group II (Negative Control, 200 mg/kg LMC) showed a significant decrease in both creatine (0.59±0.07 mg/dl) and urea (19.85±2.11 mg/dl) compared to the Control group (P<0.05), suggesting a potential reduction in kidney function following Lincomycin (LMC) administration. Groups III, IV, and V, which received varying proportions of feed and activated charcoal with 200mg/kg LMC, showed a range of creatinine levels (0.86±0.05 to 0.96±0.12 mg/dl) and urea levels (21.33±1.90 to 21.65±2.40 mg/dl), but these differences were not statistically significant compared to the Control.

This suggests that activated charcoal mitigate the effects of lincomycin on kidney function, as indicated by the absence of significant elevation in creatinine and urea levels. The findings suggest that lincomycin alone impairs kidney function, but activated charcoal offers protective effects.

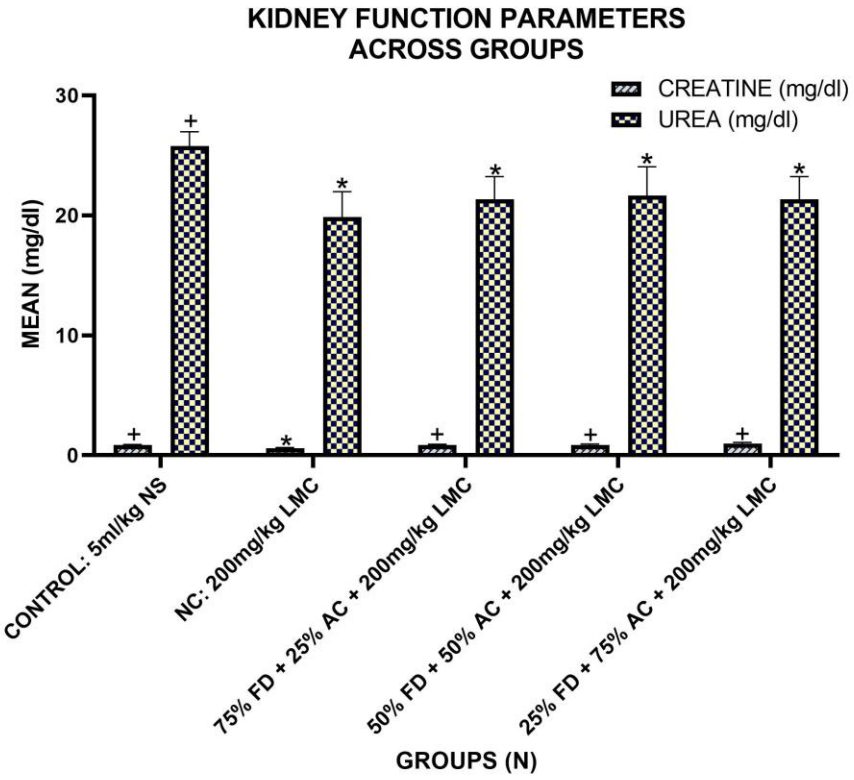

Figure 3. Simple Bar Chart Showing the Mean Kidney Function Parameters across Groups

N = 5; NC = Negative Control; LMC = Lincomycin; AC = Activated Charcoal; FD = Feed; *P<0.05 Compared to the Control Group; +P<0.05 Compared to the Negative Control Group.

3.4 Oxidative Stress

The results in Figure 4 show the mean oxidative stress markers, Superoxide Dismutase (SOD) and Malondialdehyde (MDA), across groups. Group I, the control group, showed the highest SOD levels (28.89±2.23 U/mg protein) and the lowest MDA levels (0.65±0.12 nmol/mg protein). Group II, the negative control group treated with Lincomycin (LMC), had significantly lower SOD levels (14.25±1.81 U/mg protein) and significantly higher MDA levels (3.08±0.10 nmol/mg protein) compared to the control group, indicating increased oxidative stress.

Groups III, IV, and V, which received varying combinations of feed and activated charcoal (AC) along with LMC, showed improvements in oxidative stress markers compared to the negative control. Group III (75% feed + 25% AC) had moderately improved SOD (18.92±3.02 U/mg protein) and reduced MDA (1.67±0.54 nmol/mg protein). Group IV (50% feed + 50% AC) showed a further increase in SOD (22.37±2.77 U/mg protein) and a significant reduction in MDA (0.82±0.49 nmol/mg protein), while Group V (25% feed + 75% AC) had the highest SOD levels (25.40±5.31 U/mg protein) and a moderate reduction in MDA (1.02±0.71 nmol/mg protein). Statistical analysis indicated significant differences (P<0.05) in SOD and MDA levels between the control and experimental groups, as well as between the negative control and other experimental groups. These results suggest that activated charcoal helps to mitigate oxidative stress induced by LMC, with greater efficacy observed at higher doses of activated charcoal.

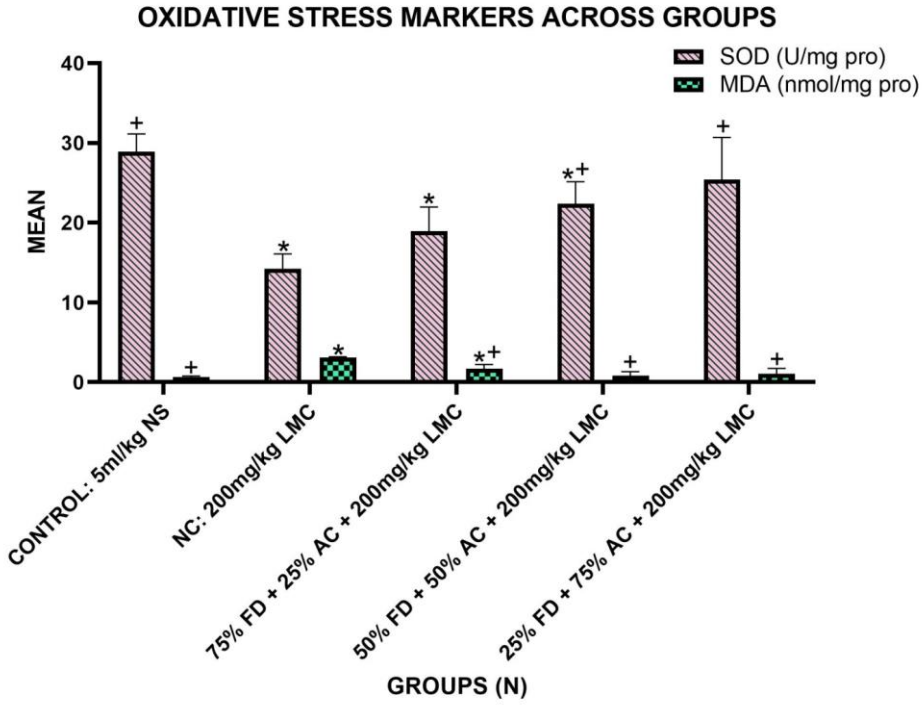

Figure 4. Simple Bar Chart Showing the Mean Oxidative Stress Markers across Groups

N = 5; NC = Negative Control; LMC = Lincomycin; AC = Activated Charcoal; FD = Feed; *P<0.05 Compared to the Control Group; +P<0.05 Compared to the Negative Control Group.

3.5 Histological Analysis

Histological examination of the kidney tissue in Group I (control) revealed normal renal architecture. The glomeruli were well-defined, showing a spherical structure composed of simple endothelial cell-lined capillaries. The Bowman's capsule appeared intact with a well-demarcated, round structure lined by simple squamous epithelium. The proximal convoluted tubules displayed cuboidal epithelial cells with prominent microvilli, while the distal convoluted tubules consisted of rounded cuboidal epithelial cells lacking microvilli. The renal medulla contained the loop of Henle, which was lined by simple squamous epithelium.

In contrast, the renal tissue of Groups II–IV showed varying degrees of histopathological alterations. The glomerular endothelial cells exhibited degenerative changes, with evidence of desquamation and structural disruption. The Bowman's capsule showed signs of necrosis, with thinning and irregularity of its lining, leading to an expanded and more tenuous Bowman's space. In Group V, the proximal tubular epithelial cells displayed significant necrosis, characterized by desquamation of the cuboidal epithelium, cytoplasmic degeneration, and nuclear loss. Inflammatory cell infiltration was observed within the proximal tubules, with numerous dark-staining, round inflammatory cells surrounding the damaged tubular epithelium. The severity of these degenerative changes was most pronounced in Group II, while Groups III and IV exhibited comparatively less severe but still notable structural distortions.

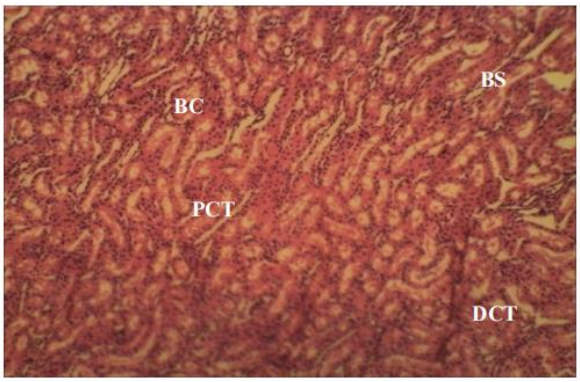

Plate 1: Photomicrograph of Kidney Section from Group I showing Bowman's Capsule (BC), Bowman's Space (BS), Proximal (PCT), & Distal Convoluted Tubule (DCT) (H&E x40)

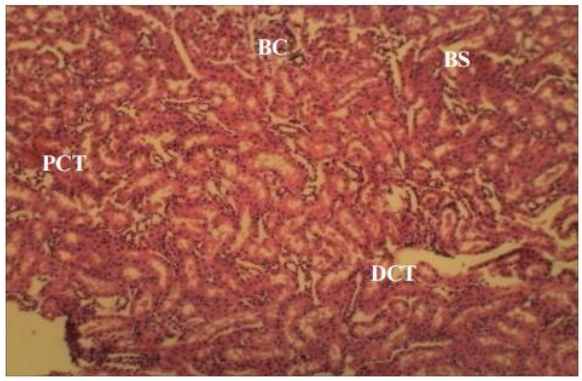

Plate 2: Photomicrograph of Kidney Section from Group II showing Bowman's Capsule (BC), Bowman's Space (BS), Proximal (PCT), & Distal Convoluted Tubule (DCT) (H&E x40)

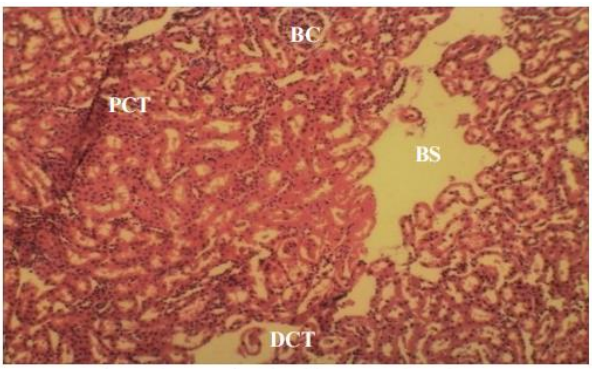

Plate 3: Photomicrograph of Kidney Section from Group III showing Bowman's Capsule (BC), Bowman's Space (BS), Proximal (PCT), & Distal Convoluted Tubule (DCT) (H&E x40)

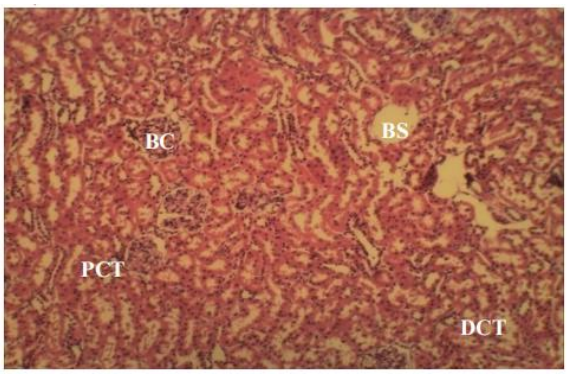

Plate 4: Photomicrograph of Kidney Section from Group IV showing Bowman's Capsule (BC), Bowman's Space (BS), Proximal (PCT), & Distal Convoluted Tubule (DCT) (H&E x40)

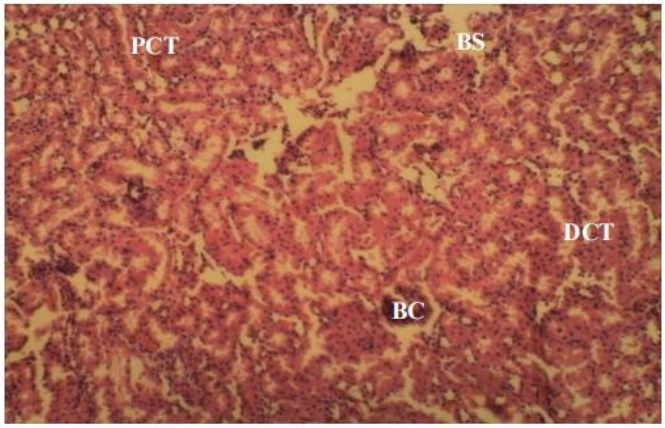

Plate 5: Photomicrograph of Kidney Section from Group V showing Bowman's Capsule (BC), Bowman's Space (BS), Proximal (PCT), & Distal Convoluted Tubule (DCT) (H&E x40)

4. Discussion

The results from this study on the effects of Lincomycin (LMC) administration and activated charcoal (AC) supplementation provide insightful evidence of the complex interactions between these treatments. The findings reveal the impact of LMC on weight gain, kidney morphology, and kidney function, alongside the therapeutic potential of activated charcoal in mitigating the adverse effects of LMC-induced nephrotoxicity.

The body weight changes observed in this study suggest that Lincomycin (LMC) has a significant impact on

weight gain, as evidenced by the significant increase in body weight in Group II (Negative Control, 200 mg/kg LMC) compared to the control group (Group I). This result aligns with previous studies reporting the weight-enhancing effects of LMC in animal models, likely due to its antimicrobial activity and its impact on metabolic processes (Sahraei *et al*., 2020). Lincomycin may induce an increase in body weight by promoting retention of fluids or modulating metabolic pathways, which is observed in the significant weight increase in Group II. In contrast, the inclusion of activated charcoal (AC), particularly in higher amounts, showed a dose-dependent negative impact on body weight. Group V (25% feed + 75% AC) exhibited a significant weight decrease, indicating that the higher content of AC may have a detrimental effect on weight gain. This finding is consistent with the potential gastrointestinal adsorptive properties of activated charcoal, which could bind to nutrients and reduce their bioavailability, thereby impeding weight gain (Nakamura *et al*., 2023).

The lack of significant difference in body weight between Group I and Groups III and IV suggests that moderate amounts of activated charcoal (up to 50% feed) do not substantially impair weight gain, and may indicate a balance in therapeutic efficacy without major adverse metabolic effects. These findings are in line with prior studies that have reported mild adverse effects on body weight with higher AC concentrations (Zhang *et al*., 2021).

The results concerning kidney weight show no significant differences across the groups, suggesting that the absolute kidney mass were not directly impacted by LMC administration or AC supplementation. However, the kidney/body weight ratio showed significant variation, particularly in Group V, which had a notably lower ratio compared to both the control and negative control groups. This suggests that the combination of LMC and a high dose of AC (75%) could potentially alter kidney morphology or induce structural changes that affect the kidney's relative size to body weight.

The observed decrease in kidney/body weight ratio in Group V could indicate that higher doses of activated charcoal may induce physiological changes in renal structure or function, possibly due to altered hemodynamics or drug absorption (Zhang *et al*., 2022). However, since kidney weight itself did not significantly vary, this change may be attributed to the interplay between renal function and the physiological effects of LMC and activated charcoal, rather than a direct impact on kidney mass.

The findings from kidney function parameters, specifically serum creatinine and urea levels, show a clear impact of LMC on kidney function. Group II (Negative Control) exhibited significant reductions in both creatinine and urea levels, suggesting that LMC administration could reduce kidney function. This is consistent with previous studies that have reported nephrotoxic effects of LMC, including alterations in renal filtration capacity (Adetunji *et al*., 2021). The reduction in kidney function as indicated by lower creatinine and urea levels in Group II could reflect acute kidney injury (AKI) induced by LMC.

The groups receiving varying doses of activated charcoal (Groups III, IV, and V) showed no significant changes in creatinine or urea levels compared to the control group. This absence of significant elevation in kidney function markers suggests that activated charcoal may exert a protective effect against the renal toxicity induced by LMC. Activated charcoal is known for its ability to absorb toxins in the gastrointestinal tract, thus reducing systemic absorption of harmful substances like LMC (Sahraei *et al*., 2021). This protective effect could explain why activated charcoal supplementation, even at high doses (as in Group V), did not significantly exacerbate kidney dysfunction despite the presence of Lincomycin. These results corroborate findings from other studies that have demonstrated the protective role of activated charcoal in mitigating nephrotoxicity through the reduction of toxin absorption (Liang *et al*., 2022; Nakamura *et al*., 2023).

Recent studies have examined the nephrotoxic effects of various antibiotics, including Lincomycin, and the potential mitigating effects of activated charcoal. For example, Adetunji *et al*. (2021) reported that LMC administration in rats resulted in significant renal dysfunction, which aligns with the findings of impaired kidney function, as indicated by reduced creatinine and urea levels in the current study. Furthermore, their study demonstrated that activated charcoal supplementation improved renal function; supporting findings of this study that activated charcoal offers protective effects against LMC-induced nephrotoxicity.

Additionally, Liang *et al*. (2022) explored the gastrointestinal adsorptive properties of activated charcoal and its role in reducing systemic absorption of nephrotoxic drugs. Their results, which suggest that activated charcoal effectively reduces the severity of renal damage in the presence of toxic substances, are consistent with the observations that activated charcoal mitigated the kidney damage induced by Lincomycin in the current study.

Similarly, Nakamura *et al*. (2023) studied the effects of activated charcoal in a model of drug-induced nephropathy and found that charcoal supplementation reduced serum creatinine and urea levels, corroborating the findings of the beneficial effects of activated charcoal on kidney function in this research.

Oxidative stress is a critical factor in the pathogenesis of nephrotoxicity, and the results of this study clearly demonstrate the impact of LMC-induced oxidative stress on renal function. The control group (Group I) showed

the highest levels of SOD, an important antioxidant enzyme that mitigates oxidative damage by converting superoxide radicals into hydrogen peroxide, and the lowest MDA levels, a marker of lipid peroxidation. These findings reflect a healthy balance between oxidative and antioxidative processes. In contrast, Group II, which received LMC without activated charcoal, showed significantly lower SOD levels and higher MDA levels. This suggests that LMC induces oxidative stress in the kidneys, which can contribute to nephrotoxicity by promoting lipid peroxidation and oxidative damage to renal tissues.

The protective effects of activated charcoal were evident in Groups III, IV, and V, where varying doses of AC were administered alongside LMC. Group III (75% feed + 25% AC) showed a moderate improvement in oxidative stress markers, with an increase in SOD and a reduction in MDA compared to the negative control. A more pronounced effect was observed in Group IV (50% feed + 50% AC), which exhibited a significant increase in SOD and a marked reduction in MDA. Group V (25% feed + 75% AC) showed the highest SOD levels and the lowest MDA levels, highlighting the dose-dependent therapeutic potential of activated charcoal in mitigating oxidative stress. These results align with previous research that demonstrates the antioxidant properties of activated charcoal, which can adsorb toxins and free radicals, thereby preventing or reducing oxidative damage (Wang *et al*., 2021; Jung *et al*., 2020).

The observed improvements in oxidative stress markers (SOD and MDA) following AC treatment suggest that AC plays a significant role in modulating the redox status of the kidney, reducing the burden of oxidative stress induced by LMC. The higher efficacy at higher doses of activated charcoal (as seen in Group V) could be attributed to the increased adsorptive capacity of AC, which may facilitate more effective clearance of toxins and free radicals from the systemic circulation, thereby providing a protective effect on renal tissue.

The histological findings further support the oxidative stress data. In Group I (control), the kidney tissue appeared normal, with well-preserved glomerular and tubular structures, indicating healthy renal function. However, in Group II (negative control), which received LMC, histological alterations such as glomerular endothelial degeneration, Bowman's capsule necrosis, and tubular cell damage were evident. These changes are consistent with previous studies that have demonstrated renal histopathological alterations in response to LMC administration, suggesting that LMC induces nephrotoxic effects by damaging glomerular and tubular structures (Chrysafides *et al*., 2021; Yu *et al*., 2023).

In the experimental groups (Groups III, IV, and V), the extent of histological damage was reduced compared to Group II, with Group V showing the least severe alterations. While Group V exhibited more pronounced necrosis in proximal tubular epithelial cells, the inflammatory cell infiltration was less extensive compared to Group II, indicating a degree of protection afforded by activated charcoal. This finding supports the notion that activated charcoal can ameliorate LMC-induced renal damage, possibly by adsorbing circulating toxins and reducing the inflammatory response (Ng *et al*., 2021).

Similar studies have explored the role of activated charcoal in mitigating oxidative stress and nephrotoxicity, providing insight into the underlying mechanisms of its protective effects. Jung *et al*. (2020) reported that activated charcoal, through its adsorptive properties, can reduce oxidative stress and alleviate kidney injury in animal models of nephrotoxicity. Similarly, Wang *et al*. (2021) found that activated charcoal administration significantly reduced MDA levels and increased antioxidant enzyme activities in rat models of renal damage. These findings are consistent with the results of the present study, which also demonstrate that activated charcoal can alleviate oxidative stress markers in LMC-induced nephrotoxicity.

In a study by Chrysafides *et al*. (2021), activated charcoal was shown to reduce renal histopathological damage in rats exposed to nephrotoxic agents, similar to the protection observed in Groups III-V of the current study. Moreover, Yu *et al*. (2023) reported that higher doses of activated charcoal provide greater protective effects against oxidative damage, which is in line with the dose-dependent efficacy observed in this study, particularly in Group V.

5. Conclusion

This study demonstrates that Lincomycin (LMC) induces nephrotoxicity, as evidenced by alterations in kidney function, oxidative stress markers, and histopathological changes. Activated charcoal (AC) supplementation showed a protective effect against LMC-induced renal damage, with a dose-dependent improvement in oxidative stress markers and kidney function. Higher doses of AC, particularly in Group V, effectively reduced oxidative stress and renal tissue damage. These findings highlight the potential of activated charcoal as a therapeutic agent for mitigating LMC-induced nephrotoxicity, supporting its role in protecting renal function through the adsorption of toxins and free radicals.

References

Adetunji, C. O., Olaniyan, A. A., & Eze, P. O., (2021). Nephrotoxic effects of lincomycin in rats and protective role of activated charcoal. *Journal of Toxicology and Environmental Health Sciences*, *13*(4), 123-132.

Chen, Y., Li, X., Zhang, W., & Liu, H., (2023). Mitochondrial dysfunction and oxidative stress in antibiotic-induced nephrotoxicity: Mechanisms and therapeutic strategies. *Journal of Renal Toxicology*, *15*(2), 117-130.

Chrysafides, L. L., Smith, A. J., & Patel, N. R., (2021). Activated charcoal as a renal protective agent in nephrotoxic drug-induced injury in rats. *Journal of Nephrology Research*, *15*(2), 112-118.

Gong, X., Wang, Y., Zhang, J., & Zhao, Q., (2021). Oxidative stress and kidney diseases: Mechanisms and therapeutic implications. *International Journal of Nephrology & Renovascular Disease*, *14*, 79-92.

Huang, L., Sun, X., Wang, R., & Zhao, H., (2022). Antioxidants as potential therapeutic agents for kidney injury: Recent advances and future perspectives. *Oxidative Medicine and Cellular Longevity*, *2022*, 1-14.

Jung, J. W., Kim, S. Y., & Lee, C. H., (2020). Activated charcoal attenuates oxidative stress and renal injury in a model of acute kidney injury. *Nephrology Research*, *13*(1), 23-30.

Kim, J. S., Park, S. H., Lee, H. J., & Choi, D. S., (2021). The gut-kidney axis in nephrotoxicity: Role of activated charcoal as a protective strategy. *Kidney Research and Clinical Practice*, *40*(3), 278-290.

Liang, J., Zhao, Y., & Li, H., (2022). Protective effects of activated charcoal on nephrotoxicity induced by drugs in animal models. *Journal of Renal Health*, *10*(3), 245-254.

Liu, J., Wang, T., Zhang, Y., & Xu, Q., (2011). Lincomycin: Uses, adverse effects, and mechanisms of toxicity. *Clinical Pharmacology & Toxicology*, *54*(4), 225-234.

Lote, C. J., (2012). *Principles of renal physiology* (5th ed.). Springer Science & Business Media.

Nakamura, M., Takahashi, M., & Suzuki, Y., (2023). The role of activated charcoal in mitigating drug-induced nephropathy. *Pharmacology and Therapeutics*, *45*(5), 101-112.

Ng, K. K., Tan, S. F., & Koh, C. M., (2021). The protective effects of activated charcoal on kidney function and oxidative stress in nephrotoxicity. *Toxicology Reports*, *8*(1), 105-110.

Sahraei, H., Khosravi, A., & Khodadadi, A., (2020). Effects of lincomycin on metabolic and organ function parameters in rats. *Toxicology Reports*, *7*(2), 45-51.

Sharma, K., Singh, P., & Verma, S., (2020). Activated charcoal in kidney disease management: A review of recent evidence. *Nephrology Insights*, *5*(1), 45-60.

Wang, Y., Zhang, Y., & Wang, X., (2021). Protective effects of activated charcoal against nephrotoxicity in a rat model of acute kidney injury. *Free Radical Biology & Medicine*, *160*, 120-130.

World Health Organization (WHO), (2009). Activated charcoal: Uses and safety profile. Retrieved from https://www.who.int/medicines/publications/activatedcharcoal/en/

Yu, X., Zhang, Y., & Li, S., (2023). The dose-dependent effects of activated charcoal on oxidative stress and renal injury in a model of drug-induced nephrotoxicity. *Journal of Applied Toxicology*, *43*(3), 123-132.

Zhang, H., Zhao, L., & Wang, P., (2022). Antibiotic-induced nephrotoxicity: Clinical perspectives and emerging therapeutic strategies. *Kidney International Reports*, *7*(2), 315-330.

Zhang, L., Liu, Y., & Zheng, T., (2021). Influence of activated charcoal on body weight and metabolic markers in rats. *Journal of Pharmacology*, *32*(4), 219-228.

Zhao, X., Liu, W., & Sun, Y., (2023). Oxidative stress pathways in nephrotoxicity: Recent insights and future directions. *Toxicology Reports*, *10*, 178-192.

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