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RESEARCH ARTICLE

Selenium mitigates Paraquat-induced hematotoxicity, oxidative stress, biochemical alterations and nephrotoxicity in male Albino rats

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A B S T R A C T

Paraquat is a widely used herbicide known for its high toxicity, posing serious health risks to humans and animals alike. Selenium, a vital trace element, supports the body's antioxidant defenses. This study explored its protective role against paraquat induced kidney damage, oxidative stress, and blood and biochemical changes in male albino rats. Forty-eight rats were divided into six groups and treated for 28 days: control, selenium only (0.5mg/kg), paraquat alone (10 and 20mg/kg) and paraquat combined with selenium. Blood tests, biochemical analyses, and kidney tissue examinations were conducted to assess the effects. Paraquat exposure significantly disrupted hematological parameters, with marked decreases in Hb, RBC, HCT, MCV, MCH, and MCHC, alongside increases in RDW-CV, WBC, neutrophils lymphocytes, monocytes, eosinophils and platelets counts ($P<0.05$), indicating anemia and systemic nephrotoxicity. Paraquat also elevated serum urea, uric acid, creatinine, HDL-cholesterol and kidney malondialdehyde levels. While reducing GSH and total antioxidant capacity. Histological analysis confirmed renal tissue damage. Co-administration of Se significantly attenuated these effects, restoring hematological and biochemical markers, reducing oxidative stress, and preserving renal architecture. Paraquat induced kidney damage is strongly linked to oxidative stress. Selenium supplementation effectively mitigated hematological, oxidative stress and renal dysfunction, highlighting its therapeutic potential in counteracting paraquat induced nephrotoxicity.

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INTRODUCTION

Herbicide contamination in soil, water, and biological systems poses significant risks to non-target organisms, including humans and animals (Bamal *et al*., 2024). Many commonly used herbicides, such as acetochlor, metolachlor, dicamba, and paraquat contain toxic active ingredients and co-formulants (Ruuskanen *et al.,* 2023). Among these, paraquat is known for its rapid action and non-selective weed control in more than 100 countries around the globe (Edo, 2022). However, its high toxicity and high-water solubility, has raised serious environmental and health concerns (Hassan *et al*., 2021). Paraquat is widely used in Pakistan's agricultural regions, particularly in the cultivation of cotton, rice, and vegetables. Although exact nationwide usage figures are limited, studies estimate that Pakistan uses over 130,000 metric tons of pesticides annually, with herbicides including paraquat accounting for around 6–7% of this total (Rashid *et al.*, 2022). A residue analysis conducted in Lahore revealed significant levels of paraquat remaining on commonly consumed produce, including guava (6.6 mg/kg), eggplant, and cluster beans. These values exceeded international safety limits, indicating unsafe application practices and posing potential risks to consumers (Abbas *et al*., 2024). The presence of such residues in fruits and vegetables sold in urban markets reflects the unregulated and excessive use of paraquat in peri-urban farming zones.

Experimental data also confirms paraquat's widespread usage across different agro-climatic zones. For

example, a field trial in Multan during 2018–2019 reported the application of paraquat-dichloride (494 IU/mL) on wheat and cotton plots to suppress weeds and boost yield performance (Khan *et al.,* 2021). Additionally, a case report from Peshawar (2022) described the death of a 3-year-old child due to accidental ingestion of paraquat stored in an unlabelled household bottle, highlighting the dangers of poor pesticide storage and inadequate medical preparedness (Shah and Aalia, 2022). These reports reveal that despite its toxicity, paraquat continues to be used extensively across Punjab, Sindh, and KPK without sufficient regulatory oversight or public awareness of its hazards.

Human exposure to paraquat may occur through skin contact, inhalation, or ingestion, with acute poisoning often leading to multi-organ failure and death (Nikdad *et al*., 2020). Paraquat-induced toxicity affects vital organs such as the lungs, liver, kidneys, heart, and immune system, with respiratory failure being a common cause of mortality (Edo, 2022). Hematological alterations, including decreased erythrocyte indices and hemoglobin levels, alongside increased packed cell volume, have also been reported following paraquat exposure (Chen *et al*., 2021). The primary mechanism of paraquat toxicity involves redox cycling, which generates reactive oxygen species (ROS) notably superoxide anions, hydrogen peroxide, and hydroxyl radicals. These ROS initiate lipid peroxidation and oxidative damage, especially in renal tissues (Idris *et al*., 2020).

Given this background, antioxidant therapy has gained attention as a potential protective strategy. Selenium, an essential trace element with strong antioxidant properties, plays a key role in maintaining redox balance, protecting cellular membranes, and preventing oxidative damage (Idris *et al*., 2020; Mustafa *et al*., 2023). Its function is primarily mediated through selenoproteins, such as glutathione peroxidase, which neutralize ROS and limit lipid peroxidation (Nunes *et al*., 2017; Hamza *et al*., 2023). Selenium has been shown to protect against chemically induced renal injury by reducing oxidative stress and preserving tissue architecture. The kidneys, which naturally store higher levels of selenium, are particularly responsive to its protective effects. This study was designed to investigate the protective role of selenium in mitigating paraquat-induced hematological, biochemical, oxidative, and histopathological alterations in male albino rats (Sharifi-Rigi and Heidarian, 2019).

MATERIALS AND METHODS

Chemical and reagent: Paraquat (Catalog No. 36541, ≥98% purity, Sigma-Aldrich, USA) was used and obtained from a local herbicide supplier in Punjab Province, Pakistan. Sodium Selenite (Na_2SeO_3, 99%, Product No.: S5261; Sigma-Aldrich, USA) purchased from a pharmaceutical company, Pakistan.

Study animals: Forty-eight healthy adult male albino rats (200–250g) were kept in the Animal House of Government College University, Faisalabad, Pakistan. Before the experiment, the animals were acclimatized for two weeks under controlled laboratory conditions (25 ± 2°C, moderate humidity, 12-hour light/dark cycle). They were housed in clean, ventilated polypropylene cages with free access to standard food and water. All procedures were approved by the Institutional Animal Ethics Review Committee, Ref. NO. GCUF/ERC/571 was ensured through daily monitoring and strict adherence to ethical guidelines.

Study design*:* After a two-week acclimatization period, the 48 rats were randomly split into six equal groups (Appendix 1). Paraquat was administered at a dose of (low dose 10mg/kg body weight and high dose 20mg/kg body weight), which is widely used to induce oxidative stress and systemic toxicity in rodent model without causing immediate lethality (Edo, 2022). This dose reliably produces sub-lethal toxicity, allowing assessment of biochemical, hematological, and histopathological changes over time.

Appendix 1: Experimental design for different treatments in the study

Sr. No.	Groups	Treatment
1	Group 1	Normal saline and standard feed
2	Group 2	Selenium@ 0.5mg/kg body weight
3	Group 3	Paraquat @10mg/kg body weight
4	Group 4	Paraquat @ 20mg/kg body weight
5	Group 5	Co-treated with paraquat (10 mg/kg) and selenium (0.5 mg/kg)
6	Group 6	Co-treated with paraquat (20 mg/kg) and selenium (0.5 mg/kg)

All treatments were given once daily by oral gavage for 28 days. Selenium was given @ 0.5mg/kg body weight each day as described by Shalaby *et al*. (2025), chosen for its well-known antioxidant and protective effects. The combination of these agents was chosen to evaluate the synergistic or additive protective effects against paraquat-induced toxicity. Co-administration allows the exploration of potential interaction effects and the capacity of the antioxidant agent to modulate oxidative stress, inflammatory responses, and tissue injury induced by paraquat. Throughout the study, the rats had free access to standard chow and fresh water.

Blood collection and hematological assessment: At the end of the treatment, blood samples were collected under mild anesthesia by cardiac puncture, using EDTA-coated tubes to prevent clotting. The samples were then analyzed for hematological parameters with an automated hematology analyzer.

The following indices were measured:

- Red Blood Cell Count (RBCs)
- Hematocrit (HCT)
- Mean Corpuscular Volume (MCV)
- Mean Corpuscular Hemoglobin Concentration (MCHC)
- Red Cell Distribution Width – Coefficient of Variation (RDW-CV)
- White Blood Cell Count (WBC)
- Differential Leukocyte Count: Neutrophils, Lymphocytes, Monocytes, Eosinophils
- Platelet Count

For biochemical analysis, blood was collected from each rat into plain tubes (without anticoagulant) and left to clot at room temperature. The clotted blood was centrifuged at 3000xg for 15 minutes to obtain serum, which was aliquoted and stored at –80°C until analysis. Following euthanasia, kidneys were excised, rinsed with 0.9% NaCl to remove blood and debris, and divided into two portions. One part was homogenized in 1.15% KCl (pH 7.4, EDTA buffer) and centrifuged at 250xg for 20 minutes to collect the supernatant for oxidative stress assessment. The second portion was fixed in 0.5% formaldehyde for histopathological evaluation.

Renal function tests estimation: Serum biochemical parameters were evaluated using commercially available enzymatic colorimetric kits (Bio Systems S.A. Barcelona, Spain; Batch No. C220134), following the manufacturer's instructions for accuracy and consistency. Assessed parameters included renal function markers (urea, creatinine, uric acid), lipid profile components (total cholesterol, HDL-C, LDL-C), and serum albumin to gauge protein status and liver function. These markers offered valuable insights into metabolic and renal changes caused by paraquat and the potential protective effects of selenium treatment.

Urea levels were determined using the urease–Berthelot method, where enzymatic hydrolysis releases ammonia that forms a blue indophenol complex, measured at 578nm. Serum creatinine was analyzed using the Jaffe reaction, producing a red-orange complex measured at 520nm. Uric acid concentrations were measured via the uricase–peroxidase method, which generates a chromogenic product also read at 520nm. Total cholesterol, HDL-cholesterol, and albumin were assessed using standard enzymatic colorimetric assays, while LDL-cholesterol was calculated using the Friedewald equation. All absorbance readings were obtained using a UV–visible spectrophotometer (BioTek ELx800, USA), and concentrations were expressed in mg/dL, following manufacturer protocols for accuracy.

Determination of some oxidative stress parameters

Determination of MDA in kidney tissues: Malondialdehyde (MDA) levels, a marker of lipid peroxidation, were measured using a modified method by Xu *et al*. (2017). Briefly, 0.1mL of sample was mixed with 0.9mL distilled water, followed by 0.5mL of 25% trichloroacetic acid (TCA) and 0.5mL of 17% thiobarbituric acid (TBA) in 0.3% NaOH. The reaction mixture was incubated in a water bath at 95°C for 40 minutes. After cooling, the absorbance was measured at 532nm using a spectrophotometer. Results were compared against a blank to determine MDA concentration.

Determination of GSH content in kidney tissues: Reduced glutathione (GSH) levels were determined following the method described by Goda *et al*. (2024). Tissue homogenates were deproteinized using 10% trichloroacetic acid and centrifuged to obtain the supernatant. For the assay, 2mL of phosphate buffer (pH 8.4) was added to each test tube, followed by 0.01mL of the supernatant. The reaction with Ellman's reagent (DTNB) produced a yellow chromophore, which was measured spectrophotometrically after vortexing and a 15-minute incubation period. GSH concentrations were calculated using a standard curve derived from the linear equation $y=mx$.

Measurement of total antioxidant activity: Total antioxidant capacity (TAC) was measured using the Ferric Reducing Ability of Plasma (FRAP) assay, as described by Benzie and Strain (1996). This method quantifies antioxidant power based on the reduction of the Fe^{3+}-TPTZ complex to a blue-colored Fe^{2+} form, with absorbance measured Spectrophotometrically. The first reagent mixes included an acetate buffer at pH 3.6 combined with ferric chloride at 10:1 concentration ratio. The experiment required tripyridyl triazine as its third component at the 1:1 ratio. Each vial of working reagent distributed 1.8 meters across three tubes that contained samples and standards and blank solutions amounting to 60µL. A mixed reaction solution underwent incubation at 37°C for 10mins before the researchers initiated the mixing process. The spectroscopic evaluation occurred at 593nm to measure the blue-colored reaction mixture. Standard solution with 1000µmol/L of ferrous sulfate along with distilled water formed the composition of the blank solution.

Histopathological analysis: Kidney tissues were fixed, dehydrated, and processed through de-alcoholization, infiltration, and paraffin embedding, following the protocol described by Gattea *et al*. (2021). Paraffin-embedded sections were cut at appropriate thicknesses and stained with hematoxylin and eosin (H&E) for histological evaluation. Microscopic examination was performed using a light microscope (Nikon Eclipse E200, Japan) at magnifications of 100X and 400X to assess morphological and cellular alterations. This histopathological analysis as described with little modification was conducted to characterize structural kidney damage induced by paraquat exposure and to determine the extent of tissue preservation or recovery afforded by selenium treatment.

Statistical analysis: Statistical analyses were conducted using GraphPad Prism version 6.0 for Windows (GraphPad Software, San Diego, CA, USA). All quantitative data are presented as mean ± standard error of the mean (SEM). Differences among experimental groups were assessed using one-way analysis of variance (ANOVA), followed by Tukey's post hoc test for multiple comparisons. A p-value of less than <0.05 was considered indicative of statistical significance.

RESULTS

Hematological analysis: Exposure to paraquat led to significant hematological disturbances, indicating systemic toxicity and hematopoietic suppression (Table 1). Both RBC count and hematocrit (HCT) parameters were significantly reduced in paraquat treated groups (G3 and G4), especially in G4 (20mg/kg), suggesting anemia and impaired erythropoiesis. Co-administration of selenium (G5 and G6) notably restored these levels, with G6 showing the most improvement, indicating selenium's protective effect against paraquat -induced oxidative damage. paraquat exposure resulted in reduced MCV and

MCHC, reflecting microcytic, hypochromic anemia. Selenium supplementation improved these indices, though the differences were not always statistically significant. An increase in RDW-CV was observed in paraquat-treated rats, indicating anisocytosis due to oxidative stress. Selenium partially normalized this variability. Total WBCs, neutrophils, lymphocytes, monocytes, and eosinophils were all significantly elevated in paraquat-only groups (G3 and G4), reflecting immune activation and inflammation. Selenium co-treatment (G5 and G6) mitigated these elevations, supporting its immunomodulatory and anti-inflammatory properties. Paraquat exposure significantly increased platelet counts, a response commonly linked to systemic stress and inflammation. Selenium helped reduce platelet levels toward normal.

Serum biochemical assays (Renal function tests): Paraquat exposure led to notable changes in several serum biochemical markers, highlighting its harmful effects, while selenium supplementation showed clear signs of protection.

Kidney function indicators: Rats exposed to paraquat had significantly higher levels of urea, creatinine, and uric acid (Fig. 1-3) compared to the control group, pointing to serious kidney damage. However, rats that received selenium along with paraquat had values much closer to normal, suggesting selenium helped protect kidney function.

Antioxidant estimation: Paraquat treated rats showed raised levels of total cholesterol, triacylglycerides, and LDL-cholesterol, along with a significant drop in HDL-cholesterol (Fig. 4-6). These changes reflect a disrupted lipid metabolism. In contrast, selenium supplementation helped lower harmful lipid levels and improved HDL-cholesterol, suggesting it helped maintain healthier blood

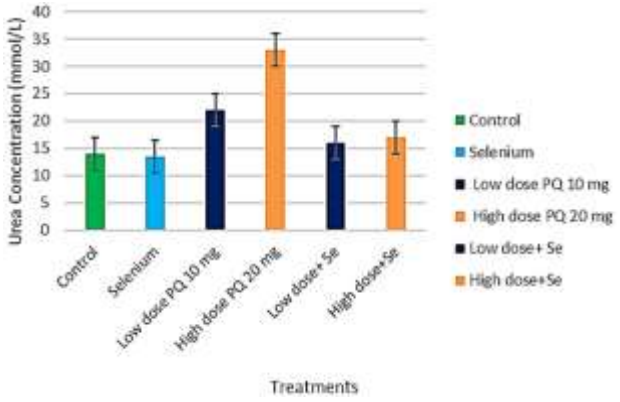

Fig. 1: Serum urea concentration (mmol/L) in male albino rats treated with paraquat, selenium and their combinations. Values are expressed as mean ± SEM (n=6), with vertical error bars representing the standard error of the mean (SEM). Paraquat-treated groups (10 and 20mg/kg) showed a significant elevation in urea levels compared to the control (P<0.05; P<0.01), indicating renal impairment. Co-administration of selenium (0.5mg/kg) resulted in a notable reduction in urea levels, highlighting its protective effect against paraquat induced nephrotoxicity.

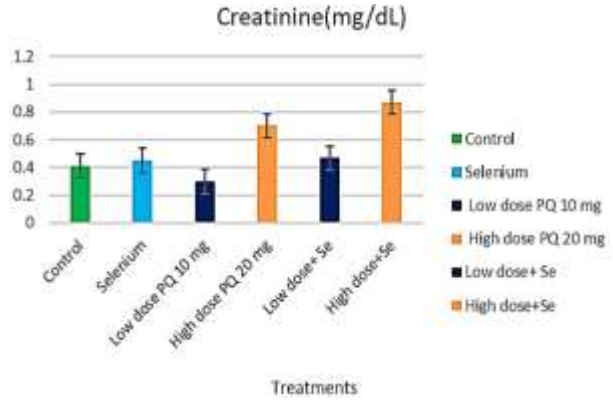

Fig. 2: Serum creatinine levels (mg/dL) in male albino rats following treatments with paraquat, selenium and their combinations. Data are expressed as mean±SEM (n=6), with vertical bars indicating the standard error of the mean. A marked increase in creatinine concentration was observed in rats exposed to high-dose paraquat (20mg/kg), suggesting renal dysfunction. Co-administration of selenium with low-dose paraquat showed a slight reduction in creatinine, while high-dose paraquat combined with selenium still exhibited elevated levels, though slightly lower than paraquat alone.

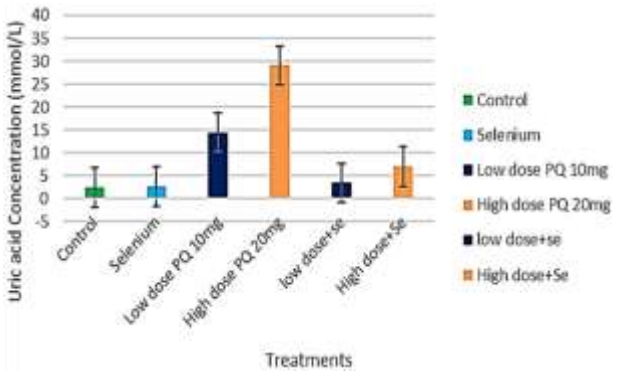

Fig. 3: Changes in serum uric acid concentration (mmol/L) in male albino rats following treatments with paraquat, selenium and their combinations. Values are expressed as mean±SEM (n=6), with vertical bars representing the standard error of the mean. uric acid levels increased significantly in rats treated with high-dose paraquat (20mg/kg), indicating oxidative stress and potential kidney damage. Co-treatment with selenium helped lower uric acid levels, particularly in the low-dose paraquat group, suggesting that selenium may offer protection against paraquat induced renal and oxidative damage.

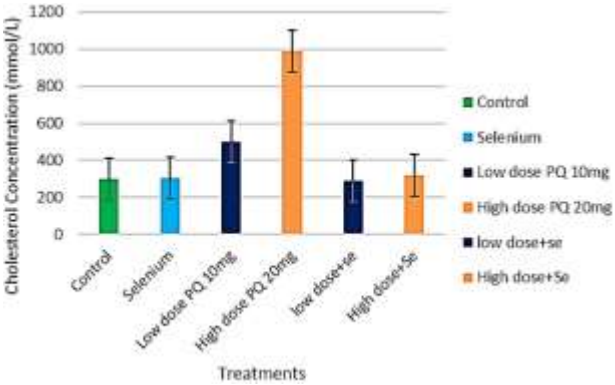

Fig. 4: Changes in cholesterol concentration (mmol/L) in male albino rats following treatments with paraquat, selenium and their combinations. Values are presented as mean±SEM (n=6). Vertical bars indicate the standard error. A significant increase in cholesterol levels was observed in rats treated with high-dose paraquat (20mg/kg), reflecting oxidative stress and potential renal damage. Co-administration of selenium notably reduced cholesterol levels, especially in the low-dose paraquat group, suggesting a protective effect against paraquat-induced renal and oxidative injury.

Table 1: Effects of selenium and paraquat, alone and in combination, on hematological parameters in male albino rats

	G1	G2	G3	G4	G5	G6
Parameters	Control	Selenium 0.5mg/kg	Paraquat 10mg/kg	Paraquat 20mg/kg	Paraquat 10mg/kg +Selenium (0.5mg/kg)	Paraquat 20mg/kg + Selenium 0.5mg/kg
RBCs ($\times10^6$/µL)	6.97±0.006***	10.95±0.005**	4.00±0.01**	3.67±0.17	6.95±0.005**	9.33±0.50**
HCT (%)	40.36±0.05*	48.76±0.05*	36.00±0.05	28.30±0.28	45.76±0.05*	40.00±0.05*
MCV (fL)	70.00±0.06	85.73±0.06(ns)	64.80±0.15*	60.13±0.38	69.73±0.07	67.70±0.08
MCH (pg)	26.56±0.05*	30.46±0.05*	18.73±0.06(ns)	15.36±0.15	25.03±0.06	17.73±0.06
MCHC (g/dL)	22.60±0.06*	28.40±0.06*	16.60±0.05*	11.70±0.06	22.10±0.05	27.10±0.05
RDW-CV (%)	14.03±0.06*	17.73±0.06*	1.26±0.05*	19.23±0.10	17.70±0.06	18.10±0.06
WBCs ($\times10^3$/µL)	9.36±0.05*	9.13±0.06*	13.53±0.05*	15.36±0.13	10.00±0.05	10.40±0.05
Neutrophils (%)	14.00±0.50	16.66±0.50**	37.66±0.50 (ns)	40.00±0.33	15.00±0.05	18.00±0.06
Lymphocytes (%)	47.33±0.69*	46.33±0.83*	88.66±0.50 (ns)	90.66±0.50	71.00±0.06	80.00±0.50
Monocytes (%)	2.00±0.30**	3.00±0.00	9.66±0.50**	18.66±0.50	3.00±0.072	4.00±0.07
Eosinophils (%)	4.00±0.00 (ns)	9.00±0.00 (ns)	15.00±0.00 (ns)	16.00±0.00	2.00±0.30	2.50±0.02
Platelet Count ($\times10^3$/µL)	410±0.66	500±0.50	650±0.50*	700±0.50	420±0.50	450±0.50

Values are expressed as mean ± SEM (n=6). treatment groups: G1– Control, G2 – Selenium (0.5mg/kg), G3 – paraquat (10mg/kg), G4 – paraquat (20mg/kg), G5 – paraquat(10mg/kg) + Selenium (0.5mg/kg), G6 – paraquat (20mg/kg) + Selenium (0.5mg/kg). Statistical significance compared to control: *P<0.05, **P<0.01, ***P<0.001, ns = not significant.

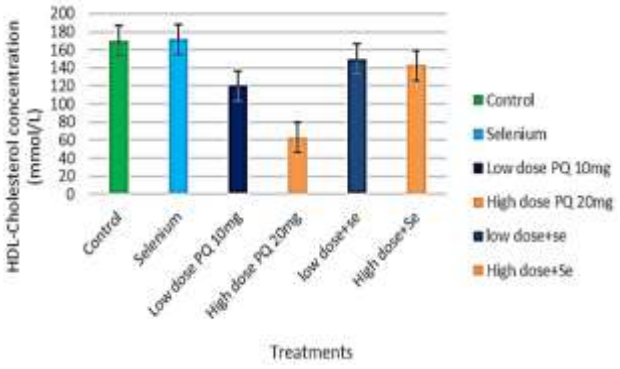

Fig. 5: Changes in High-density lipoprotein (HDL) cholesterol concentration (mmol/L) in male albino rats following treatments with paraquat, selenium and their combinations. Values are expressed as mean±SEM (n=6), with vertical bars representing the standard error of the mean. levels of high-density lipoprotein **(**HDL) cholesterol concentration decreased significantly in rats treated with high-dose paraquat (20mg/kg), indicating oxidative stress and potential kidney damage. Co-treatment with selenium helped normal the high-density lipoprotein cholesterol concentration levels, particularly in the low-dose paraquat group, suggesting that selenium may offer protection against paraquat induced renal and oxidative damage.

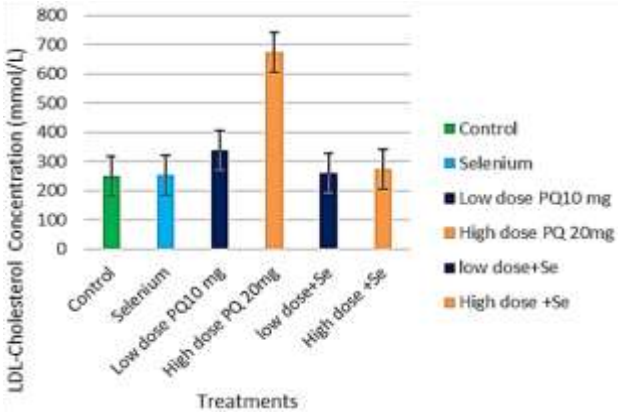

Fig. 6: Changes in low-density lipoprotein (LDL-cholesterol) concentration (mmol/L) in male albino rats following treatments with paraquat, selenium and their combinations. Values are mean±SEM (n=6). High-dose paraquat significantly raised LDL cholesterol levels, indicating oxidative stress and kidney damage. Selenium co-treatment, especially at lower paraquat doses, helped normalize LDL levels, highlighting its protective potential.

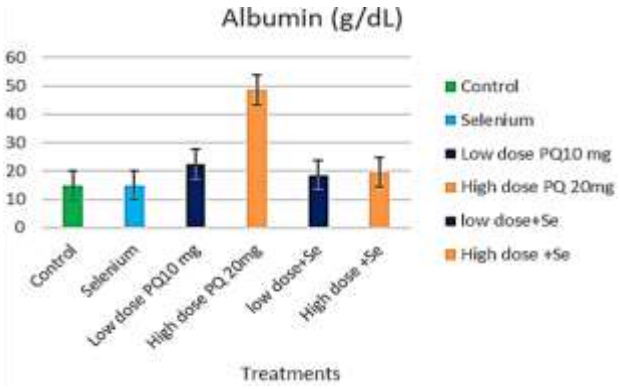

Fig. 7: Changes in albumin concentration (g/dL) in male albino rats following treatments with paraquat, selenium and their combinations. Values are mean ± SEM (n=6). High-dose paraquat significantly increased albumin levels, indicating kidney damage and oxidative stress. Selenium co-treatment, especially at low paraquat doses, helped restore albumin levels, suggesting a protective effect.

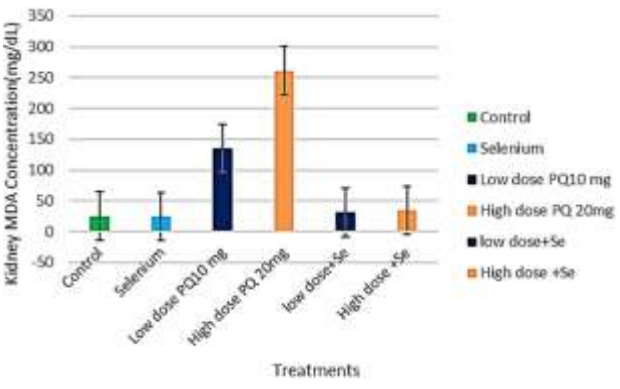

Fig. 8: Serum levels of malondialdehyde (MDA) levels (mg/dL) in male albino rats following treatments with paraquat, selenium and their combinations. Data are mean ± SEM (n=6). Paraquat significantly increased MDA levels (P<0.05), indicating oxidative stress. Selenium co-treatment reduced MDA, suggesting antioxidant protection.

lipid levels. Paraquat caused a dose-dependent rise in LDL-cholesterol levels in rats, with the high dose showing severe disruption. However, selenium co-treatment significantly reduced these levels, highlighting its protective role in maintaining lipid balance in (Fig. 6).

Serum albumin: Albumin levels a marker of protein status and kidney function were significantly reduced in paraquat-exposed rats (Fig. 7). This decline was reversed in selenium-treated animals, further supporting its protective role.

Oxidative stress parameters: Experimental animals treated with paraquat exhibited significantly elevated kidney malondialdehyde levels compared to the control group (P<0.05), as shown in Fig. 8. Administration of supplements effectively reduced malondialdehyde (MDA) concentrations in paraquat treated animals, resilient statistically significant improvements (P<0.05). As illustrated in Fig. 9, the paraquat group showed decreased levels of reduced glutathione (GSH) relative to controls. However, co-administration dose of paraquat with selenium resulted in a significant (P<0.05) increase in renal glutathione (GSH) content. Paraquat exposure significantly reduced kidney TAC levels, especially at the high dose, reflecting elevated oxidative stress. In contrast, selenium co-treatment noticeably restored TAC levels, bringing them closer to those in control and selenium-only groups. This highlights selenium's protective role in maintaining antioxidant balance in renal tissue as illustrated in Fig. 10.

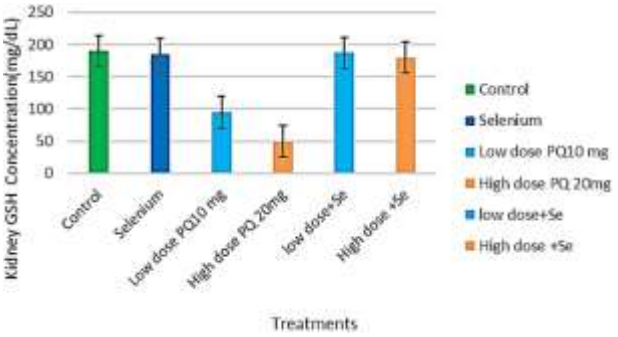

Fig. 9: Serum levels of malondialdehyde (GSH) levels (mg/dL) in male albino rats following treatments with paraquat, selenium and their combinations. Values are expressed as mean ± SEM (n=6), with vertical bars representing standard error. Paraquat reduced glutathione (GSH) levels in a dose-dependent manner, while selenium alone elevated GSH. Co-treatment with selenium significantly restored GSH levels, confirming its antioxidant protective effect.

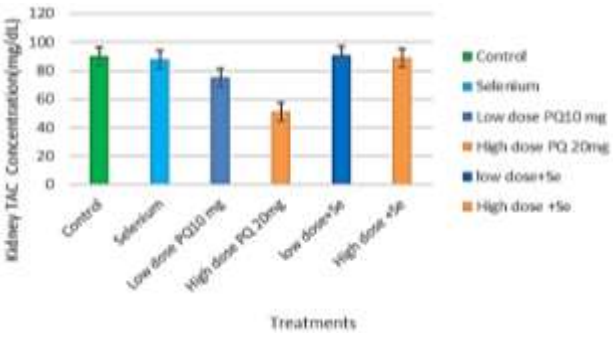

Fig. 10: Serum levels kidney total antioxidant capacity (TAC) levels (mg/dL) in male albino rats following treatments with paraquat, selenium and their combinations. Values are expressed as mean ± SEM (n=6), with vertical bars indicating standard error. Paraquat exposure caused a dose-dependent decline in total antioxidant capacity (TAC). However, selenium co-treatment significantly restored TAC levels toward normal, indicating its protective antioxidant role.

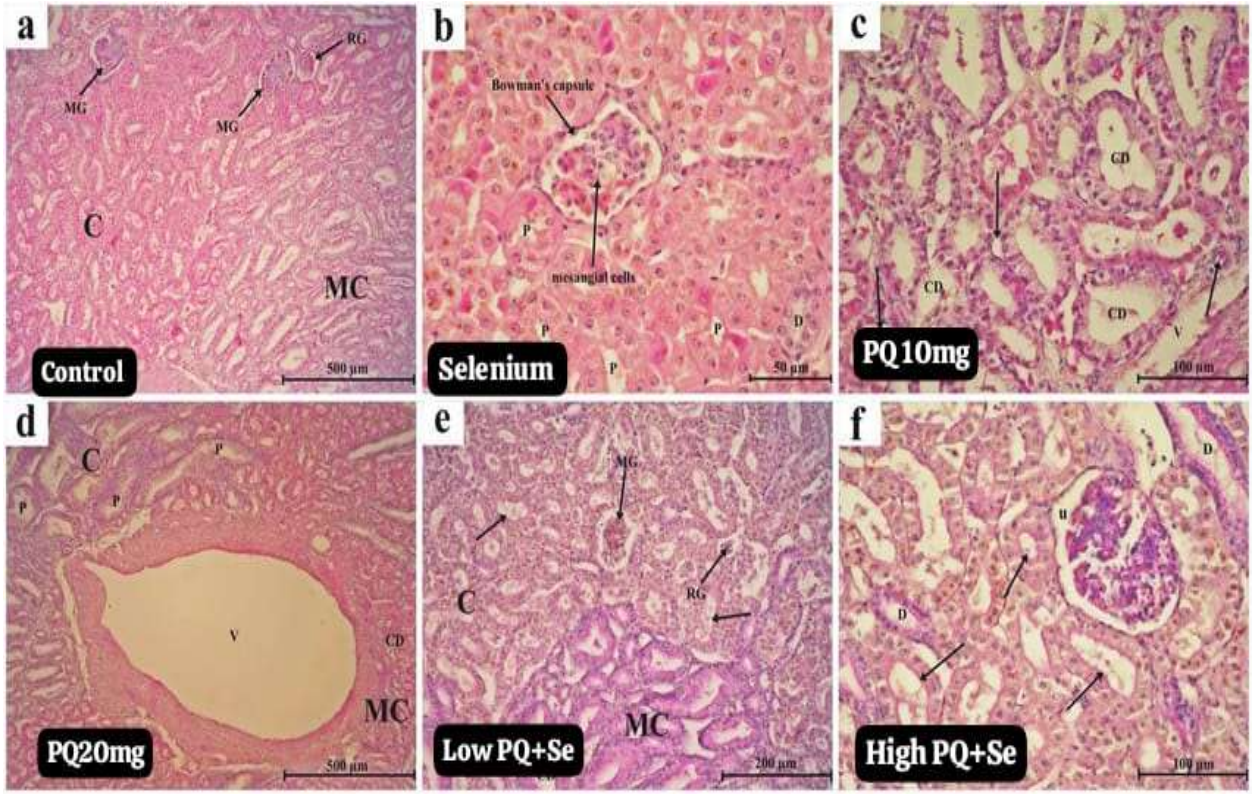

Fig. 11: The histological comparison of kidney tissues under different experimental conditions, stained with Hematoxylin and Eosin (H&E) through 400X magnification between each treatment group. Each panel (a–f) represents kidney tissue under various treatments. (a) Control: Normal renal structure with intact glomeruli (RG, MG), cortex (C), and medullary ducts (MC). (b) Selenium: Normal glomerular and tubular architecture; visible Bowman's capsule, proximal (P) and distal (D) tubules. (c) paraquat 10mg/kg: Tubular degeneration and vascular damage (CD, V). (d) paraquat 20mg/kg: Severe structural damage, dilated vessels (V), and disrupted tubules (P, D). (e) Low paraquat+Selenium: Partial recovery; preserved glomeruli (RG, MG) and collecting ducts (MC). (f) High paraquat+Se: Improved morphology with defined distal tubules (D) and reduced urinary space (u), indicating selenium's protective effect.

Histopathological findings of kidney tissue: Kidney tissue sections stained with hematoxylin and eosin (H&E) were examined under a light microscope at 400X magnification (Fig. 11). The control and selenium-only groups showed normal renal histo-architecture with well-preserved glomeruli, Bowman's capsules, and intact proximal and distal tubular epithelium, indicating no adverse effects from selenium supplementation. Paraquat exposure induced marked pathological changes, including expansion of Bowman's space, tubular epithelial desquamation, necrosis, and focal hemorrhage. The distinction between proximal and distal tubules was disrupted due to extensive epithelial shedding. Co-administration of selenium with paraquat showed partial improvement in renal morphology. Although some structural disruptions such as widened Bowman's space and minor hemorrhages persisted, epithelial damage was reduced compared to paraquat only groups. The low-dose paraquat with selenium group exhibited relatively better renal architecture, with clearly defined tubules and fewer necrotic areas, suggesting a protective effect of selenium against paraquat induced nephrotoxicity.

DISCUSSION

This study explored the damaging effects of paraquat and how selenium might help reduce these effects. Paraquat is a commonly used herbicide that causes harm by generating reactive oxygen species (ROS), leading to oxidative stress and injury to body tissues (Kheiripour *et al*., 2021; Aboubakr *et al*., 2023). In our study, paraquat exposure led to a marked drop in red blood cells, hematocrit, MCV, and MCHC, indicating the development of microcytic hypochromic anemia. These changes suggest that paraquat may suppress bone marrow function or cause red cell destruction, as also reported in earlier research (Wajda *et al*., 2020). The increase in RDW-CV and white blood cells further supports that paraquat triggers an inflammatory response and blood cell abnormalities. Interestingly, when selenium was given alongside paraquat, blood parameters improved significantly, highlighting selenium's role in fighting inflammation and oxidative damage (Shalaby *et al*., 2025). Renal function markers were also significantly altered upon paraquat exposure, with elevated serum urea, creatinine, and uric acid levels indicating renal dysfunction. These biochemical changes reflect nephrotoxicity consistent with the histopathological observations of tubular necrosis, glomerular atrophy, and interstitial inflammation. The renal impairments observed are consistent with prior reports of paraquat-induced oxidative renal injury (Song e*t al*., 2019; Sahu *et al*., 2020). These changes point to impaired kidney function, which aligns with histological findings of damaged tubules and glomeruli (Kheiripour *et al*., 2021). Selenium helped restore kidney function and structure, likely through its antioxidant properties and its role in supporting enzymes like glutathione peroxidase (Hou *et al*., 2021).

The study also revealed changes in lipid levels. Paraquat-treated rats had high levels of cholesterol, triglycerides, and LDL, and lower HDL levels, reflecting disturbed lipid metabolism due to liver oxidative stress. Selenium helped improve these values, probably by protecting lipids from oxidation and supporting normal liver function (Sun *et al*., 2024; Shalaby *et al*., 2025). Paraquat-treated rats showed high MDA levels and low GSH and TAC, indicating oxidative damage and weakened antioxidant defenses. Various studies (Xu *et al*. 2017; El-Boshy *et al*., 2019; Hu *et al*. 2019) have shown that paraquat may reduce the activity of antioxidant enzymes in renal tissues, which is consistent with the findings of this study. Selenium supplementation reversed these effects, reducing oxidative stress by restoring antioxidant balance.

Histological analysis confirmed paraquat's harmful impact on kidney tissues—tubular degeneration, cell shedding, and glomerular damage were clearly visible. These alterations in renal architecture led to impaired blood filtration and electrolyte imbalance in the body (See *et al*., 2022). Elevated levels of oxidative stress are the major factors to various histopathological impairments as reported earlier (Edwards and Kurtcuoglu, 2022; Men *et al*., 2022). However, selenium treatment reduced tissue damage and helped preserve the kidney's normal structure, likely due to its antioxidant and cell-protective roles (Idris *et al*., 2020). In summary, selenium showed strong protective effects against paraquat-induced blood, kidney, and oxidative stress damage. These results suggest selenium could be a useful agent in reducing herbicide-related toxicity and preserving organ function.

Conclusions: This study clearly demonstrated that paraquat induces significant kidney damage in rats by generating reactive oxygen species, including hydroxyl radicals, hydrogen peroxide, and superoxide anions. These lead to oxidative stress, lipid peroxidation, and depletion of cellular antioxidants, as reflected by elevated malondialdehyde (MDA) levels and reduced total antioxidant capacity (TAC). Selenium supplementation showed a notable protective effect by enhancing antioxidant defenses, reducing oxidative damage, and restoring serum biochemical parameters. These findings suggest that selenium holds promise as a therapeutic agent to counteract paraquat-induced toxicity. Overall, our results highlight the potential of selenium in mitigating oxidative stress and supporting renal health during herbicide exposure. Future studies are recommended to explore its long-term protective effects and validate these findings across different models.

Conflict of interest: The authors declare that there is no conflict of interest.

Regulatory compliance: All procedures followed national and institutional ethical standards for the care and use of laboratory animals.

Ethical approval: The study was approved by the Institutional Animal Ethical Review Committee under approval: Ref. NO. GCUF/ERC/571

Authors' contribution: MUG and AR Conceptualization, Supervision, critical review, editing; KMA and AA resource supplies in kind and histopathology; MRS and SH conducted the experiments; SK and BH formal and statistical data analysis; QUA manuscript preparation and laboratory analysis.

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