Original Article

CoQ10 Attenuates Atrazine-Induced Hepatotoxicity: A Histological and Biochemical Study

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ABSTRACT

Objective: The study aims to elucidate the impact of oxidative stress on liver function and to explore the therapeutic potential of CoQ10 in mitigating this herbicide-induced hepatic toxicity.

Study Design: Experimental study

Place and Duration of Study: This study was conducted at the Anatomy department of Islamic International Medical College in cooperation with National Institute of Health (NIH) in Islamabad from July 2023 to June 2024.

Methods: Thirty adult male Sprague Dawley rats weighing between 200 and 250 g were split into three groups at random: Group A was the control group, Group B was the illness group that received atrazine treatment, and Group C was the intervention group that received both atrazine and CoQ10. As indicators of liver damage, alkaline phosphatase (ALP), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) were examined. To evaluate structural alterations, liver samples were also examined histopathologically.

Results: Atrazine exposure significantly increased oxidative stress and reduced antioxidant enzyme activities, resulting in elevated liver enzyme levels and significant hepatocellular damage. CoQ10 supplementation mitigated these effects by reducing oxidative stress, restoring antioxidant enzyme activity, and normalizing liver enzyme levels ($p < 0.05$). Histopathological analysis revealed marked improvement in liver architecture with CoQ10 treatment.

Conclusion: CoQ10 demonstrate strong hepatoprotective effects against atrazine-induced oxidative damage by reducing liver injury and restoring antioxidant defense mechanisms. These findings suggest its potential as a therapeutic agent for managing atrazine-induced hepatic injury.

Key Words: Atrazine, Hepatotoxicity, Coenzyme Q10, Antioxidant, Liver Injury

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INTRODUCTION

The triazine family is extensively utilized for controlling both broadleaf and grassy weeds. Atrazine, a widely applied member of this family, is commonly used in crops such as corn, sugarcane, and sorghum to enhance agricultural growth and productivity.[1,2] Despite its numerous benefits, the widespread use of atrazine has sparked environmental and human health related concerns. Its remarkable stability and persistence in the environment have led to significant bioaccumulation in soil and freshwater bodies, including rivers, lakes, and even rainwater. This contamination has caused severe aquatic pollution, harming non-target organisms long after its initial application.[3,4]

Atrazine's ubiquity is troubling due to its well documented adverse effects. It has been shown to disrupt endocrine systems causing reproductive and developmental issues as well so poses significant health risks to humans and other mammals.[5]

The primary cause of atrazine's toxicity is thought to be its ability to cause oxidative stress. Reactive oxygen species (ROS), which can harm biological constituents like lipids, proteins, and nucleic acids, are produced in greater quantities when atrazine is exposed. Because of its high metabolic rate and mitochondrial composition, the liver, body's main detoxifying organ, is especially susceptible to oxidative damage. The emergence of liver illnesses is facilitated by this oxidative stress, which interferes with basic cellular processes.[6,7]

Coenzyme Q10 (CoQ10) is a naturally occurring compound with antioxidant properties and a key role in cellular energy production. It participates in the mitochondrial electron transport chain, facilitating ATP synthesis, the primary energy currency of the cell. Beyond its role in energy metabolism, CoQ10 is a

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potent antioxidant, capable of scavenging free radicals and regenerating other antioxidants such as vitamin E.[8,9]

CoQ10 has been extensively studied for its antioxidant effects and its protective role against oxidative stress and toxicity across various models.[10] It has been shown to mitigate oxidative damage caused by environmental pollutants, pharmaceutical drugs, and radiation. Studies have demonstrated that CoQ10 supplementation alleviates hepatic injury in animal models of drug-induced liver damage.[11] Furthermore, CoQ10's protective effects extend beyond the liver, shielding vital organs such as the heart and kidneys from oxidative stress, suggesting its broad-spectrum antioxidant potential.[12]

This article reviews prior research on toxicity and highlights the protective effects of CoQ10 on atrazine-induced liver damage, as these effects were still not explored. The study aims to elucidate the impact of oxidative stress on liver function and to explore the therapeutic potential of CoQ10 in mitigating this herbicide-induced hepatic toxicity. These findings could contribute to public health strategies, particularly for populations exposed to atrazine through agricultural practices or contaminated water supplies.

METHODS

Chemicals and Reagents: Atrazine, in a 38% suspension formulation and COQ10 in powder form was used in this experiment.

Animal Model and Housing: After receiving university ethical approval (Riphah/IRC/1070), the experiment was carried out in the anatomy department of Islamic International Medical College in cooperation with the National Institute of Health (NIH) in Islamabad. Total 30 male Sprague Dawley rats weighing 200–250 grams and 8–11 weeks of age were used in this investigation. The rats were kept in cages of 40 by 40 by 60 cm, and they were given water and rat pellets to eat. The rats were kept in a 12-hour light-dark cycle with 50% humidity at a standard temperature of 22 ± 0.5°C.[13,14]

Grouping: The rats were divided into 3 groups, 10 rats in each group.

Group A (Normal control): Rats in this group were fed a standard pellet chow diet without any treatment.

Group B (Disease control): Rats were administered atrazine orally at a dose of 120 mg/kg body weight daily for 21 days.[15]

Group C (Intervention group): Rats were treated with atrazine at a dose of 120 mg/kg body weight orally, along with Coenzyme Q10 (CoQ10) supplementation at 120 mg mixed into their chow daily for 21 days.[10]

After 21 days of treatment, the rats were euthanized, dissected and their livers, after removal, immediately washed with cold saline fixed in formalin.

Sampling: Blood samples were obtained using anticoagulated microcapillary syringes, allowed to coagulate for 30 minutes at room temperature, and then centrifuged at 3000 rpm prior to dissecting the retro-orbital venous plexus. Pure sera that had not been hemolyzed were immediately separated and kept at -20°C for biochemical examination. As directed by the manufacturer, commercial diagnostic kits (BioAssay Systems) were used to measure the serum levels of liver enzymes, including alkaline phosphatase parameters, aspartate aminotransferase, and alanine amino-transferase.[14]

Histopathological Examination: For histopathological examination, liver tissues were fixed in 10% buffered formalin for 48 hours. The fixed tissues were subsequently dehydrated using graded concentrations of ethanol, cleared with xylene, and embedded in paraffin wax. Serial sections, 5 µm thick, were prepared using a microtome and stained with hematoxylin and eosin (H&E). These stained sections were then examined under a light microscope to assess histopathological changes in liver tissue architecture.

Statistical Analysis: Statistical analyses were performed using statistical package for social sciences (SPSS) version 26 and a P-value of less than or equal to 0.05 was considered significant. Data was presented as mean ± SD. Quantitative variables were analyzed by one way ANOVA, followed by Tukey´s post hoc test for between-group comparisons while qualitative variables were analyzed by Chi Square test.

RESULTS

Liver Function Tests: Serum levels of liver enzymes (ALT, AST, and ALP), were measured to assess liver function. The atrazine-treated group experienced a significant increase in ALT levels (82.4 ± 7.1) compared to the control group (35.6 ± 4.3), indicating liver damage. However, in the group treated with both atrazine and CoQ10, ALT levels (48.7 ± 5.2) were significantly reduced and were comparable to the control group, highlighting CoQ10's hepato-protective effect (table 1).

Similarly, AST levels were significantly higher in the atrazine-treated group compared to the control group, but the addition of CoQ10 led to a notable reduction in AST levels, further demonstrating CoQ10's protective effect on liver function. Additionally, ALP levels were significantly elevated in the atrazine-treated group, reflecting liver damage. The group treated with both atrazine and CoQ10 showed a significant reduction in ALP levels compared to the atrazine group, reinforcing the hepatoprotective role of CoQ10.

Table No.1:Comparison of biochemical findings among different groups

GROUPS	ALT Mean ± SD (IU/L)	P-Value	AST Mean ± SD (IU/L)	p-value	ALP Mean ± SD (IU/L)	p-value
A	35.6 ± 4.3		105.2 ± 6.1		85.7 ± 7.3	
B	82.4 ± 7.1	**0.04***	148.6 ± 12.4	**0.03***	162.4 ± 15.2	**0.001****
C	48.7 ± 5.2	**0.001****	104.3 ± 8.9	**0.001****	115.2 ± 10.8	**0.001****

Analysis of Histopathology: In the liver sections of the control group, a single layer of hepatocytes formed the hepatic cords around the compact central veins. The hepatocytes had granular cytoplasm, evenly distributed around large, centrally positioned, well-stained spherical nuclei. The nuclei featured distinct nucleoli and chromatin material. Classic portal triads or tetrads were embedded in connective tissue. Binucleated hepatocytes were less common than mononucleated ones (Figure 1A). In the groups treated with CoQ10 (figure 1C) displayed a normal appearance like the control group. However, mild changes were observed in the atrazine (120 mg/kg) group, including slight congestion of the portal vein and sinusoids (1B). Hepatocyte vascular congestion was categorized based on the extent of involvement in the evaluated fields. The hepatic damage was categorized as; none (0) indicated no observable damage, mild (1) represented damage affecting 0–25%, moderate (2) 25–50%, and severe (3) indicated damage affecting 50–100%.of of the liver architecture.

In group A, 10% of rats showed mild congestion and 90% showed no congestion. In group B, 30% of rates showed mild congestion, 30% severe while 40% showed moderate congestion. In group C, 40% exhibit mild congestion while 60% showing normal liver architecture in hepatic lobule. The Chi-square analysis revealed a highly significant difference among the experimental groups, with a *p-value* of less than 0.001 (table 2). In the control group, rats did not exhibit any sinusoidal dilatation. In group B, 60% showed moderate dilatation, while 10% had severe sinusoidal dilatation. For group C, 30 % of the rats exhibited mild dilatation , and 70% displayed normal liver lobule . The Chi-square analysis indicated a highly significant difference among the experimental groups, with a p-value of less than 0.001.

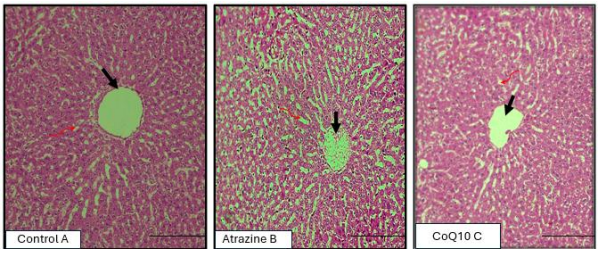

Figure No.1: Histopathological findings of different groups. Black arrow showing vascular congestion and red arrow showing sinusoidal dilatation.

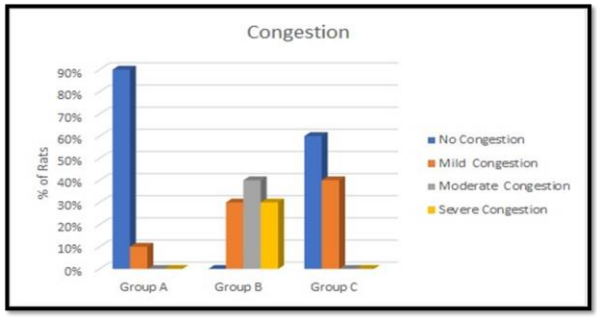

Figure No.2: Bar chart showing % of rats in degree of hepatocyte congestion

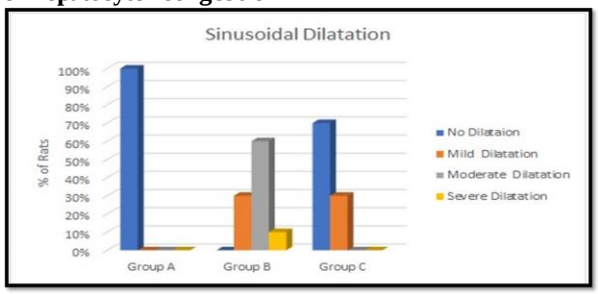

Figure No.3: Bar chart showing% of rats in degree of sinusoidal dilatation

DISCUSSION

The current study offers strong proof of Coenzyme Q10's (CoQ10) ability to shield rats' livers against atrazine-induced damage. A common herbicide, atrazine poses several health hazards because of its environmental persistence and capacity for bioaccumulation. Its hepatotoxic effects, primarily mediated through oxidative stress, have been well-documented. Atrazine exposure is known to elevate oxidative stress markers, disrupt the antioxidant defense system, and cause liver congestion, sinusoidal dilatation, and apoptosis.[1,16] Our results suggested that CoQ10 effectively counteracts atrazine-induced oxidative damage by enhancing the liver's antioxidant capacity.

Significant hepatic damage is indicated by elevated blood liver enzyme values, such as ALT, AST, and ALP, seen in the group treated with atrazine. These results are in line with earlier studies by Muhammad *et al*., which found that atrazine exposure resulted in a significant rise in liver enzymes, which is indicative of oxidative stress and inflammation-induced hepatic cell damage.[17] Similarly, Abarikwu *et al*. highlighted that liver enzyme elevation is a hallmark of hepatic dysfunction caused by environmental toxins such as

atrazine, which compromises membrane integrity and disrupts normal cellular processes.[18]

CoQ10 supplementation in the present study significantly mitigated this enzyme elevation, restoring liver function tests closer to the normal values. This underscores CoQ10's potent hepatoprotective effects, likely mediated through its antioxidant and anti-inflammatory properties. Saleh and also demonstrated CoQ10's capabilities to ameliorate hepatic damage against Acrylamide-induced oxidative stress. Elshazly *et al* also studied the beneficial effects of this supplement on the liver in 2020. Their findings support the notion that CoQ10 reduces hepatocellular injury.[19,20] Another researcher further reported that CoQ10 enhances antioxidant enzyme activities, contributing to the restoration of tissue integrity and function. Our histopathological findings reinforce these biochemical observations by revealing marked disturbances. Rashad et al also found this type of correlation between liver enzymes elevations and liver architecture damage induced by atrazine.[21] Muhammad et al also observed this pattern of atrazine induced hepatotoxicity in their experiment.[17] Atrazine induced hepatic damage was successfully treated with CoQ19 in our experiment. Wang and his colleagues explained the hepatoprotective effects of this enzyme in a recent study. They concluded that these protective effects are mediated through its antioxidant properties.[22]

CONCLUSION

The current study offers strong proof of Coenzyme Q10's (CoQ10) ability to shield rats' livers against atrazine-induced damage.

This might have significant effects on the development of treatment plans to stop atrazine-induced liver damage, particularly in communities exposed to high concentrations from contaminated water sources or agricultural practices. CoQ10 supplementation may serve as a therapeutic remedy against atrazine-induced alterations in health.

Future recommendation: Further studies are needed to identify the exact molecular mechanism of action by which CoQ10 exerts its protective effects and in other atrazine-related target organs with special attention on dose-dependency thus deserved long-term efficacy. Clinical trials should be conducted to investigate the protective effects of CoQ10 on atrazine-induced liver toxicity in humans, exploring optimal dosage and duration of supplementation.

Author's Contribution:

Concept & Design or acquisition of analysis or interpretation of data:	Sadia Farooq, Shabana Ali, Arifa Haroon
Drafting or Revising Critically:	Afifa Siddique, Tayyaba Qureshi, Tayyaba Fahad
Final Approval of version:	All the above authors
Agreement to accountable for all aspects of work:	All the above authors

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