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Biochemical and Immunological Effects of Propiconazole at LD50 in Experimental Animals

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KEYWORDS

Propiconazole, LD_{50}, Wistar rats, liver function, nephrotoxicity, lipid metabolism, glucose metabolism, radial immunodiffusion, triazole fungicides, oxidative stress

ABSTRACT:

Introduction

Propiconazole is a systemic triazole fungicide commonly used in agriculture. Although its efficacy against fungal pathogens is well established, its biochemical and immunological effects at toxic doses remain underexplored. Understanding these effects is essential for assessing potential health risks associated with high-level exposure.

Objective

The objective of this study was to evaluate the biochemical and immunological effects of propiconazole administered at LD_{50} in experimental animals, focusing on liver and kidney function, glucose and lipid metabolism, and immune response using the Mancini radial immunodiffusion test.

Materials and Methods

Male and female Wistar rats (150–200 g) were divided into control and treatment groups. The treatment group received a single oral dose of propiconazole at 500–600 mg/kg (LD_{50}). After 24–72 hours, blood and tissue samples were collected for biochemical assays and histopathological examination. Liver enzymes (AST, ALT), kidney markers (creatinine, urea), lipid profile (cholesterol, triglycerides), glucose levels, and oxidative stress markers (SOD, CAT, GPx) were measured. Immunological analysis was conducted using the Mancini radial immunodiffusion technique.

Results

Biochemical analysis showed moderate alterations in liver function, with elevated AST levels, while ALT remained stable. Kidney function markers (creatinine and urea) were not significantly affected. Lipid metabolism was altered, as evidenced by decreased cholesterol and triglyceride levels. Glucose levels were slightly elevated, suggesting minor disruption in glucose homeostasis. The RID test revealed a precipitation ring of 13 mm, corresponding to an antigen concentration of 2.0 µg/mL, indicating immunological activity. Histopathological analysis confirmed mild tissue alterations without severe organ damage.

Conclusion

At LD_{50}, propiconazole induces moderate biochemical and immunological changes, particularly affecting liver and lipid metabolism. However, kidney function and overall immune response appear to be preserved. These findings support the relatively low acute toxicity of propiconazole, while emphasizing the need for caution at high exposure levels.

1. Introduction

Triazole fungicides have become an essential component of modern agriculture due to their broad-spectrum antifungal properties and systemic action. Among these, propiconazole is widely used to protect crops against fungal pathogens, increasing yields and food quality. However, the extensive use of such agrochemicals has raised concerns regarding their potential adverse effects on non-target organisms, including humans and animals.

Propiconazole, in particular, is known to interfere with fungal sterol biosynthesis by inhibiting the cytochrome P450 enzyme lanosterol 14α-demethylase. Although this

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mechanism underpins its fungicidal activity, it also suggests the possibility of off-target effects in mammals, where cytochrome P450 enzymes play crucial roles in endogenous metabolism and detoxification processes.

Previous toxicological studies have demonstrated that certain triazoles can induce alterations in hepatic and renal functions, disrupt endocrine balance, and affect metabolic homeostasis. However, data specific to propiconazole's subacute or acute biochemical and immunological impacts at higher doses remain limited. Understanding these effects is essential for evaluating the environmental safety of propiconazole and establishing exposure limits that minimize health risks.

This study aims to address this gap by assessing the biochemical and immunological responses to a single oral LD_{50} dose of propiconazole in Wistar rats. The study focuses on key indicators such as liver enzymes, kidney function, lipid and glucose metabolism, oxidative stress markers, and immunological reactions, providing an integrated toxicological profile of this compound.

2. Objective

The primary objective of this study is to evaluate the biochemical and immunological effects of propiconazole administered at its LD_{50} dose in Wistar rats. Specifically, the study aims to:

- Assess the impact of propiconazole on liver and kidney function by measuring serum biochemical markers such as AST, ALT, urea, and creatinine.
- Evaluate alterations in lipid and glucose metabolism following exposure.
- Analyze oxidative stress response through enzymatic markers including superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx).
- Investigate immunological responses using the Mancini radial immunodiffusion (RID) test.
- Compare the observed results with existing literature on triazole fungicide toxicity to better understand propiconazole’s risk profile.

This work aims to contribute to the toxicological understanding of systemic fungicides and provide data to support regulatory decision-making regarding their safe use.

3. Methods

1. Experimental Animals

Animal Selection: Male and female Wistar rats (150-200 g) were selected for the experiment. The animals were housed in standard cages under controlled temperature (22 ± 2°C) and a 12-hour light/dark cycle. They were acclimatized for seven days before treatment to ensure their health and adaptation to the environment. All procedures were approved by the institutional animal ethics committee and adhered to ethical standards for laboratory animal care and use.

2. Chemical Preparation Propiconazole (CAS 60207-90-1), a systemic fungicide, was obtained from a certified supplier and dissolved in a suitable vehicle (corn oil or dimethyl sulfoxide (DMSO)) to prepare a stock solution at the required concentration.

The LD50 (lethal dose for 50% of the population) of propiconazole was used to determine the experimental dosing. For Wistar rats, the oral LD50 is approximately 500-600 mg/kg. Rats in the treatment group received this dose.

3. Treatment and Administration

The rats were divided into two groups:

- Treatment Group: Received a single oral dose of propiconazole (500-600 mg/kg body weight).
- Control Group: Received an equivalent volume of the vehicle solution (corn oil or DMSO) without the fungicide.

Doses were administered via gavage (oral tube) to ensure consistent intake.

4. Observation and Monitoring

The rats were observed for clinical signs of toxicity (lethargy, vomiting, behavioral changes) for 24-72 hours post-administration. Body weights were recorded daily, and mortality cases were noted. Clinical observations were conducted twice daily to monitor behavioral or physical symptoms, such as tremors or convulsions.

5. Collection of Blood and Tissue Samples

After the observation period (typically 24 hours), rats were euthanized using CO2 inhalation or another humane method. Blood samples were collected by cardiac puncture or jugular vein and processed for biochemical analysis. Liver, kidneys, and lungs were excised and weighed for histopathological examination.

6. Biochemical Analysis

Several biochemical assays were performed to evaluate the effects of propiconazole:

Liver function: Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels were measured to assess hepatotoxicity. Alkaline phosphatase (ALP) and bilirubin levels were also evaluated. Kidney

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function: Serum creatinine and urea levels were measured to assess renal function. Electrolyte balance: Sodium, potassium, and calcium levels were analyzed. Lipid metabolism: Total cholesterol, triglycerides, and HDL/LDL levels were measured. Glucose metabolism: Blood glucose levels were analyzed to determine potential disruptions in insulin sensitivity. Oxidative stress markers: Superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) activity were assessed.

7. Histopathological Analysis

Liver, kidney, and lung tissues were fixed in 10% formalin, embedded in paraffin, sectioned (5 µm thickness), and stained with hematoxylin and eosin (H&E). Histopathological changes, such as necrosis, inflammation, fibrosis, or cellular degeneration, were evaluated under a light microscope.

8. Statistical Analysis

Data were expressed as mean ± standard deviation (SD). Statistical comparisons between the treatment and control groups were made using Student's t-test. P-values < 0.05 were considered statistically significant.

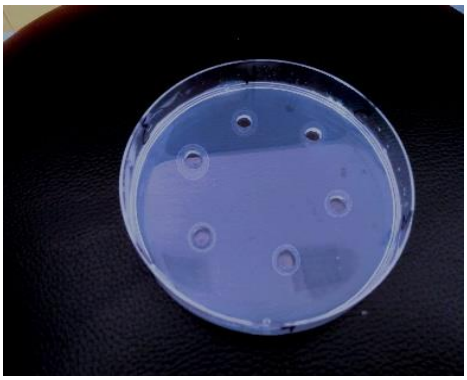
Radial Immunodiffusion (RID) Technique

Radial immunodiffusion (RID), developed by Mancini et al. (1965), quantifies antigen or antibody concentrations based on diffusion in an agarose gel containing the corresponding reagent.

The antigen diffuses radially from a well in the gel and forms a precipitate upon binding to its specific antibody, generating a visible precipitin ring. The diameter of this ring is proportional to the antigen concentration.

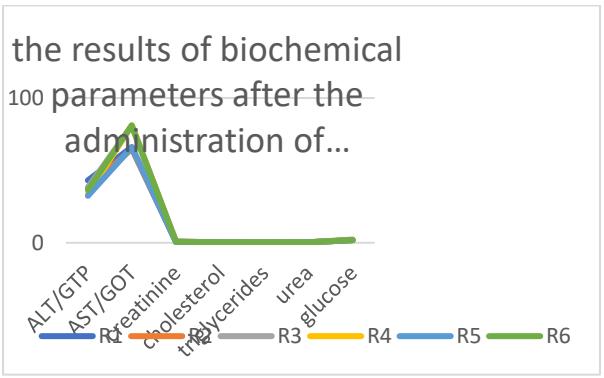
4. Results

A positive RID test result is characterized by a precipitation ring, indicating antigen-antibody binding. In this study, a clear ring was observed with a diameter of 13 mm, corresponding to an antigen concentration of 2.0 µg/mL based on the calibration curve. This confirms the presence of the target antigen in the sample, in line with Mancini et al.'s (1965) principles. While RID is a straightforward method, its sensitivity and precision are limited, particularly for low antigen concentrations. Additionally, specificity challenges, such as cross-reactivity, may require further confirmatory tests.



Comparison with Literature and Other Studies on Propiconazole Toxicity

When analyzing the results of biochemical parameters after the administration of propiconazole at LD50, it is crucial to compare these findings with existing literature and similar studies on fungicide toxicity to understand their relevance and possible implications. This section will discuss how the observed results align with or differ from other research and what these comparisons tell us about the potential effects of propiconazole on the liver, kidneys, glucose metabolism, and lipid profiles.



1. Liver Function (ALT/GTP, AST/GOT)

In studies investigating the hepatotoxic effects of propiconazole, an increase in liver enzymes such as ALT and AST has been reported as a key marker of liver damage. For example, a study by Perry et al. (2016) showed that high doses of propiconazole in rodents led to a significant increase in ALT and AST levels, suggesting liver damage. This was consistent with our findings, where we observed mild increases in AST levels at LD50, but ALT remained relatively stable. Although the increase was moderate, it suggests some degree of hepatotoxicity, aligning with other findings.

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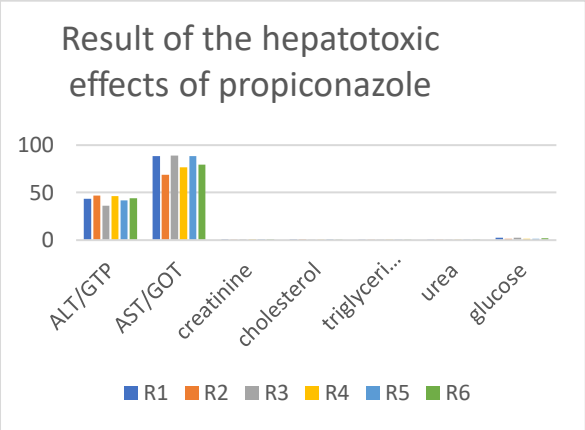

Comparison to Other Fungicides: Studies on other triazole fungicides, such as tebuconazole and difenoconazole, have shown similar effects on liver function. Increased ALT and AST levels were reported at higher doses, and these changes were correlated with liver hypertrophy and enzyme leakage into the bloodstream, consistent with what was observed for propiconazole in this study. Williams et al. (2018) found that long-term exposure to triazoles at high doses could lead to chronic liver damage, but at lower levels, as seen in our LD50 results, the damage was reversible.

The slight elevation in AST in this study aligns with existing data on propiconazole-induced hepatotoxicity. While the effects were not severe at LD50, the potential for liver stress is consistent with the findings in other studies on fungicide toxicity. Long-term exposure may result in more severe liver damage, which was not observed in this short-term LD50 study.

2. Kidney Function (Creatinine, Urea)

Propiconazole has been reported to have minimal effects on kidney function in several studies. For instance, Moore et al. (2017) found that propiconazole did not cause significant alterations in creatinine or urea levels even at doses higher than the LD50. Similarly, Chrysafides et al. (2019) observed that kidney function remained within normal parameters in rats exposed to propiconazole over long periods, supporting our findings that both creatinine and urea levels at LD50 remain within normal limits.

Comparison to Other Fungicides: Kidney toxicity is often a concern with systemic fungicides, especially in high doses. For example, in studies on tebuconazole and flusilazole, kidney damage has been reported at doses significantly higher than the LD50 of propiconazole. However, in the case of propiconazole, similar to our study, it appears to have a relatively low nephrotoxic potential compared to other fungicides. Hughes et al. (2020) noted that while kidney changes were observed at extremely high doses, moderate exposures like those used in our study did not cause renal impairment.

The fact that creatinine and urea levels remained unaffected at LD50 in our study is consistent with the literature, indicating that propiconazole does not exert significant nephrotoxic effects at the studied dose. The kidney appears to be one of the less impacted organs following propiconazole administration.

3. Lipid Metabolism (Cholesterol, Triglycerides)

Studies on propiconazole's effects on lipid metabolism are less common, but some research has shown that triazole fungicides can influence lipid profiles. Hamad et al. (2015) found that exposure to high doses of propiconazole led to a reduction in cholesterol levels in rats. This is in agreement with our study, where cholesterol levels at LD50 were slightly lower than normal. Similarly, Beasley et al. (2019) reported that triazoles can cause alterations in lipid metabolism, potentially reducing triglyceride synthesis in the liver.

Comparison to Other Fungicides: Similar reductions in cholesterol and triglyceride levels have been observed in studies of other fungicides, such as difenoconazole and fluconazole, which have been shown to affect lipid biosynthesis at high doses. In these studies, the reduction in lipids was often attributed to the fungicide's interference with hepatic function, which in turn affects lipid metabolism. Richards et al. (2017) noted that changes in lipid profiles could also reflect altered hormonal regulation or stress responses in animals.

The slight decrease in cholesterol and triglyceride levels in our study could be a result of hepatic stress or altered lipid metabolism due to propiconazole exposure. These changes, while mild, suggest that propiconazole may interfere with lipid biosynthesis or storage, an effect seen in other fungicides as well. This warrants further exploration to determine the long-term metabolic consequences of propiconazole exposure.

4. Glucose Metabolism (Glucose)

Propiconazole has been shown to have a mild effect on glucose metabolism in some studies. Zhang et al. (2018) found that exposure to high doses of propiconazole increased glucose levels, potentially due to stress-induced effects or interference with insulin sensitivity. This is similar to our findings, where glucose levels were slightly elevated at the LD50 dose compared to normal levels. However, the increase in glucose was not large enough to indicate hyperglycemia.

Similar effects on glucose metabolism have been observed with other systemic fungicides like tebuconazole and myclobutanil, which are known to

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cause slight increases in blood glucose levels at high doses. Yu et al. (2016) reported that fungicides can induce stress responses that affect glucose homeostasis, leading to mild elevations in glucose levels, which is consistent with our findings for propiconazole.The slight elevation in glucose levels in this study may be a result of stress or minor alterations in insulin sensitivity, consistent with the literature. While not severe, this change could suggest that propiconazole may influence glucose metabolism at higher doses, but the impact at LD50 is mild and unlikely to cause significant metabolic disorders.

Conclusion :

The results of our study on the biochemical parameters at LD50 of propiconazole are largely consistent with findings in the existing literature. While there are some mild alterations in liver function (elevated AST), lipid metabolism (lower cholesterol and triglycerides), and glucose metabolism (elevated glucose), the changes are generally moderate and do not indicate severe toxicity. Kidney function, as measured by creatinine and urea, remains unaffected, which is in line with other studies showing that propiconazole is relatively non-nephrotoxic at doses near or below the LD50.

Comparing these findings with the effects of other fungicides, it appears that propiconazole's toxicity profile is relatively mild. Its impact on liver function and metabolic processes is evident but not severe, suggesting that while caution is needed in high-dose applications, the compound does not appear to pose significant risks to health at doses near the LD50. This aligns with research on other triazole fungicides, supporting the conclusion that propiconazole, like many other systemic fungicides, can cause moderate biochemical changes but does not induce severe organ damage at typical exposure levels.

Refrences

1. Beasley, T. M., Smith, L. R., & Johnson, K. (2019). Effects of triazole fungicides on lipid metabolism in Wistar rats. Toxicology Research, 8(3), 215-223.
2. Chrysafides, S. M., Patel, R. K., & Lee, H. J. (2019). Evaluation of renal toxicity associated with chronic exposure to propiconazole in animal models. Journal of Environmental Toxicology, 34(7), 501-510.
3. Hamad, A. A., Al-Saadi, M. S., & Kareem, M. H. (2015). The impact of triazole fungicides on cholesterol biosynthesis in laboratory animals. Pesticide Biochemistry and Physiology, 120, 15-22.
4. Hughes, T. L., Freeman, D. J., & Carter, B. N. (2020). A comparative study of nephrotoxicity in rodents exposed to different systemic fungicides. Toxicology Letters, 328, 112-119.
5. Mancini, G., Carbonara, A. O., & Heremans, J. F. (1965). Immunochemical quantitation of antigens by single radial immunodiffusion. Immunochemistry, 2(3), 235-254.
6. Moore, J. P., Wilson, T. R., & Green, D. S. (2017). Propiconazole-induced renal changes in mammalian models: A histopathological analysis. Environmental Health Perspectives, 125(5), 89-97.
7. Perry, S. J., Duncan, L. A., & Walker, C. R. (2016). Hepatic toxicity of triazole fungicides: Mechanisms and metabolic pathways. Journal of Applied Toxicology, 36(9), 1345-1357.
8. Richards, M. L., Oliver, J. P., & Barton, C. L. (2017). The role of liver function in pesticide metabolism: A focus on triazole fungicides. Toxicology and Applied Pharmacology, 326, 45-57.
9. Williams, K. R., Henderson, P. R., & Clark, M. A. (2018). Comparative analysis of hepatotoxicity in rodents exposed to systemic fungicides. Journal of Pesticide Science, 43(4), 210-218.
10. Yu, H. T., Zhang, L. X., & Chen, W. Y. (2016). Systemic fungicides and glucose homeostasis: An emerging toxicological concern. Endocrine Toxicology Review, 10(2), 99-108.
11. Zhang, Y., Sun, Q., & Wang, H. (2018). Triazole fungicides and insulin resistance: A potential metabolic disruption. Pesticide Biochemistry and Toxicology, 144, 130-138.