

Hepatoprotective Effects of *Camellia Sinensis* Against Tartrazine-Induced Liver Toxicity in Albino Rats

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ABSTRACT

This study was designed to evaluate the hepatotoxic effects of tartrazine and to examine the protective potential of *Camellia sinensis* (green tea) in albino rats. Exposure to tartrazine led to noticeable alterations in liver function markers, as reflected by elevated levels of ALT, AST and ALP, which indicate liver damage. Histopathological observations further supported these findings, showing degeneration of hepatocytes, necrosis, and inflammatory cell infiltration in liver tissues. These changes are mainly linked to oxidative stress caused by excessive generation of reactive oxygen species.

On the other hand, administration of *Camellia sinensis* showed a clear protective effect. The treated group exhibited improvement in biochemical parameters along with restoration of normal liver architecture. This protective action is largely attributed to the antioxidant properties of green tea polyphenols, especially catechins, which help in reducing oxidative stress and maintaining cellular integrity.

Overall, the findings of the study suggest that although tartrazine may induce toxic effects on the liver, natural antioxidants such as green tea can play an important role in reducing such damage and may have therapeutic relevance in managing chemically induced hepatotoxicity.

Keywords- Tartrazine, Hepatotoxicity, *Camellia sinensis*, Tartrazine, Liver enzymes, Antioxidant, Oxidative stress.

I. INTRODUCTION

Tartrazine (E102), also known as FD & C Yellow No. 5, is a synthetic azo dye widely used as a coloring agent in food, beverages, pharmaceuticals, and cosmetics. It is primarily bright yellow and is found in a large number of different products that include soft drinks, candy, ice cream, sauces and processed foods. Tartrazine's low cost and highly stable form make it one of the most widely used food additives in the world. In recent years, however, scientific evidence continues to emerge concerning its potential toxicological effects and safety on both human and animal health. Tartrazine is metabolized in the gastrointestinal (GIT) tract through a reductive cleavage of its azo bond (i.e., via the cleavage of the azo bond, it produces aromatic amines which have been shown to have mutagenic as well as carcinogenic properties). As these metabolites (i.e., mutagenic/carcinogenic aromatic amines) enter the

blood circulation and accumulate in vital organs (particularly the liver and kidney), they cause toxic effects within those organs. Since the liver is the primary organ that detoxifies and metabolizes drugs and chemicals, it is extremely susceptible to damage from chemicals. There are several studies which indicate that exposure to tartrazine results in the following adverse effects on the liver: hepatic insufficiency (due to oxidative stress), oxidative stress, and changes in biochemical and hematological parameters (Amin *et al.*, 2010; Elbanna *et al.*, 2017). Oxidative stress is believed to be a major factor in how tartrazine causes injury to the liver. The creation of reactive oxygen species (ROS) disrupts the balance of various cellular processes, leading to damage of membranes made of lipids and proteins, as well as to DNA and ultimately to cellular injury and necrosis. Studies have shown that Chronic exposure to tartrazine has been shown to increase levels of liver enzymes in the blood (such as ALT, AST and

ALP) and therefore indicates liver injury (Golli *et al.*, 2016; Shakoor *et al.*, 2022). When experimental animals were exposed to azo dyes, many histopathological changes were seen, including degeneration of hepatocytes, inflammation and necrosis. More recently, researchers have begun exploring whether natural antioxidants might be able to protect against the effects of synthetic chemicals. One such natural antioxidant is *Camellia sinensis* (green tea) and it has attracted considerable attention because of the high content of various types of bioactive compounds, including catechins, flavonoids, polyphenols, tannins, and vitamins. EGCG is the most potent antioxidant amongst all these compounds. EGCG scavenges free radicals, inhibits lipid peroxidation, and increases the endogenous antioxidant defense mechanism of the cells. Based on a number of studies, it has been reported that various classes of toxicity are prevented through green tea consumption. Liver function is improved, and oxidative stress is decreased, as well as inflammation modifications through the consumption of polysaccharides found in green tea (Abdel-Raheem *et al.*, 2010; Vieira *et al.*, 2011).

Additionally, polyphenols found in green tea help to increase the activity of antioxidant enzymes, such as superoxide dismutase (SOD), catalase, and peroxidase of glutathione, thus reducing the damage to tissues from free radicals (Hsu *et al.*, 2014). These characteristics make *Camellia sinensis* a good candidate for the treatment of hepatotoxicity caused by tartrazine. Despite increasing awareness regarding the deleterious effects of food additives, the use of tartrazine continues to be high, especially in children and adolescents. Therefore, there is a pressing need to develop appropriate and effective strategies to mitigate the adverse effects of tartrazine on the human body. The present study was designed to investigate the hepatotoxic effects of tartrazine and to evaluate the protective role of *Camellia sinensis* through biochemical, oxidative stress, and histopathological assessments.

II. MATERIALS AND METHODS

1. Experimental Animals

Healthy adult albino rats (*Rattus norvegicus*), weighing 150–200 g, were used in this study. The animals were obtained from a certified breeding facility and acclimatized for 7 days prior to experimentation. They were housed in clean polypropylene cages under standard laboratory conditions (25 ± 2°C temperature, 50–60% humidity, and 12 h light/dark cycle). Rats were provided with standard pellet diet and water *ad libitum*. Bedding was changed regularly to maintain hygiene. Animals were randomly divided into experimental groups, and only healthy individuals were selected. All procedures were conducted following ethical guidelines for laboratory animal care and use.

2. Sample Size

Sample size was determined using statistical power analysis with a power of 0.80 and significance level (α) of 0.05 to ensure reliability of results. Based on this calculation, each experimental subgroup consisted of six animals ($n = 6$), providing adequate statistical strength and minimizing experimental variability.

3. Ethical Approval

The experimental protocol was reviewed and approved by the Institutional Animal Ethics Committee (IAEC) in accordance with CPCSEA guidelines, ensuring ethical care, handling, and use of laboratory animals.

4. Experimental Design

A total of 200 rats were divided into four groups ($n=50$) and each group was further subdivided based on duration

- Group I: Control
- Group II: Tartrazine-treated
- Group III: Green tea-treated
- Group IV: Tartrazine + Green tea

Each group was subdivided based on duration of exposure (7, 15, 30, 45, 60 days).

5. Dose Selection

- Tartrazine dose was selected as **one-tenth of LD₅₀**, based on previously reported toxicological studies to ensure sub-lethal exposure.
- *Camellia sinensis* extract dose was chosen according to **standard hepatoprotective doses** documented in earlier experimental research.

6. Green Tea Standardization

- The *Camellia sinensis* extract was standardized to contain **40–50% epigallocatechin gallate (EGCG)**, the principal bioactive compound responsible for antioxidant activity.
- The extract exhibited **apurity of ≥95%**, ensuring consistency, quality, and reproducibility of experimental results.

7. Preparation of Chemicals

Tartrazine solutions were prepared in distilled water at appropriate concentrations (7.5 mg/kg body weight). Green tea extract was prepared by steeping the leaves in hot water to obtain 10% aqueous extract.

8. Biochemical Analysis

Liver function parameters analyzed:

- ALT (Alanine aminotransferase) by Reitman and Frankel method (1957)
- AST (Aspartate aminotransferase) by Reitman and Frankel method (1957)
- ALP (Alkaline phosphatase) by Kind and King method (1954)
- Total protein by Biuret method (Gornall *et al.*, 1949)
- Bilirubin by Jendrassik and Grof method (1938)

9. Statistical Analysis

Data were expressed as mean $\pm$ standard deviation (SD). Statistical analysis was performed using one-way ANOVA followed by Tukey’s post hoc test. Differences were considered significant at $p < 0.05$.

III. RESULTS

The experimental study revealed that tartrazine exposure caused significant hepatotoxicity in albino rats.

Serum biochemical parameters such as ALT, AST, ALP and total bilirubin were markedly elevated in the tartrazine-treated group compared to control. This indicates liver cell damage and membrane leakage.

In contrast, the *Camellia sinensis*-treated group showed near-normal values, confirming its protective role. The combined treatment group (tartrazine + green tea) demonstrated partial recovery, suggesting ameliorative effects of green tea.

Table 1: Liver Function Parameters

Group	ALT (U/L)	AST (U/L)	ALP (U/L)	Total Protein (g/dL)	Bilirubin
Control	35 $\pm$ 2	40 $\pm$ 3	80 $\pm$ 4	6.5 $\pm$ 0.3	0.6 $\pm$ 0.05
Tartrazine	90 $\pm$ 5***	100 $\pm$ 6***	160 $\pm$ 8***	4.2 $\pm$ 0.2***decreased	1.8 $\pm$ 0.1***
Green Tea	38 $\pm$ 3	42 $\pm$ 4	85 $\pm$ 5	6.3 $\pm$ 0.3	0.7 $\pm$ 0.06
Combined	55 $\pm$ 4**	65 $\pm$ 5**	110 $\pm$ 6**	5.8 $\pm$ 0.2**increased	1.0 $\pm$ 0.08**

Statistical Significance Note

Values are expressed as Mean $\pm$ SD (n = 6). Statistical analysis was performed using one-way ANOVA followed by Tukey’s post hoc test.

- $p < 0.05 \rightarrow *$ (significant)
- $p < 0.01 \rightarrow **$ (highly significant)
- $p < 0.001 \rightarrow ***$ (very highly significant)

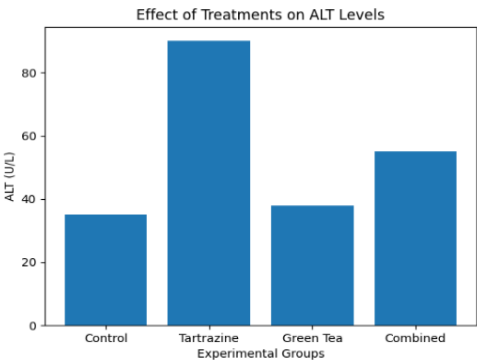


Figure 1. Effect of tartrazine and *Camellia sinensis* on ALT levels in albino rats. Tartrazine significantly increased ALT, while green tea showed protective effects.

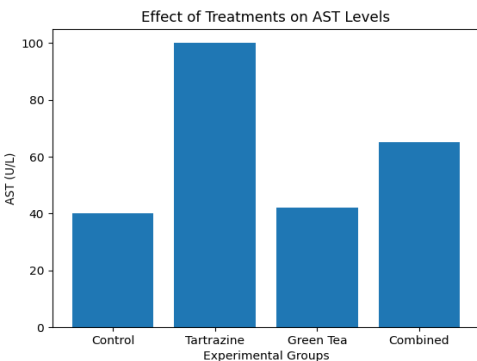


Figure 2. Effect of tartrazine and *Camellia sinensis* on AST levels in albino rats. Elevated AST in treated groups indicates hepatic stress, which was reduced by green tea.

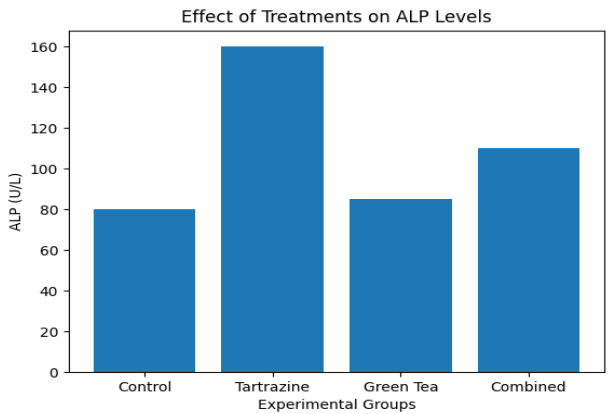


Figure 3. Effect of tartrazine and *Camellia sinensis* on ALP levels in albino rats. Green tea treatment mitigated the tartrazine-induced rise in ALP levels.

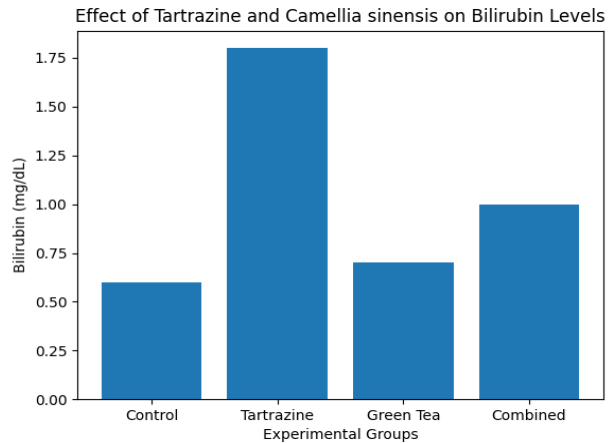


Figure 4. Effect of tartrazine and *Camellia sinensis* on serum bilirubin levels in albino rats. Tartrazine exposure significantly increased bilirubin levels, indicating hepatic dysfunction, while green tea treatment showed a protective effect by reducing bilirubin levels toward normal.

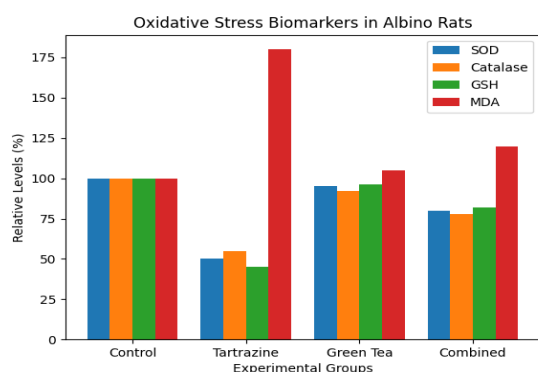


Figure 5. Effect of tartrazine and *Camellia sinensis* on oxidative stress biomarkers in albino rats. Tartrazine significantly decreased antioxidant enzymes (SOD, catalase, GSH) and increased MDA levels, while green tea treatment restored antioxidant status and reduced lipid peroxidation.

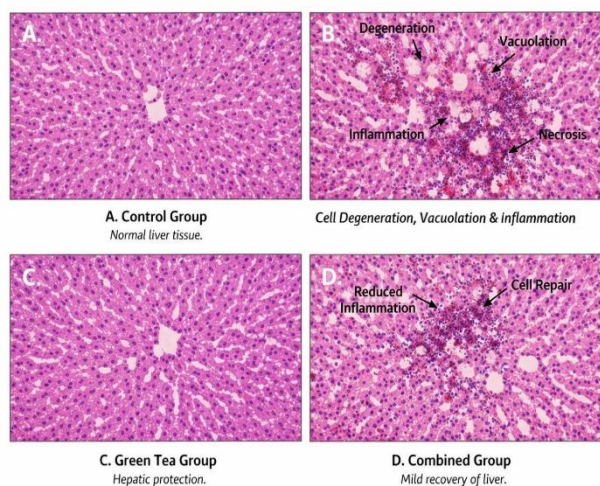


Figure 6. Histopathological changes in liver tissues of albino rats following exposure to tartrazine and treatment with *Camellia sinensis* (H&E staining, 400×).

- (A) Control group showing normal hepatic architecture with well-organized hepatocytes and intact central vein.
- (B) Tartrazine-treated group showing severe hepatocellular degeneration, cytoplasmic vacuolation, inflammatory cell infiltration, and necrosis.
- (C) *Camellia sinensis*-treated group exhibiting normal liver structure with preserved hepatocytes indicating no toxicity.
- (D) Combined treatment group (tartrazine + green tea) showing reduced inflammation, mild cellular damage, and signs of hepatic recovery.

IV. DISCUSSION

Oxidative stress is a primary mechanism underlying tartrazine-induced hepatotoxicity. The

observed elevation in liver enzymes, including alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP), indicates hepatocellular damage and increased membrane permeability. These enzymatic alterations are well-established biomarkers of liver injury and are consistent with previous reports (Amin *et al.*, 2010; Golli *et al.*, 2016). Histopathological examination in the present study revealed hepatocellular degeneration, necrosis, and inflammatory cell infiltration, confirming structural liver damage following tartrazine exposure. The toxic effects of tartrazine are largely attributed to oxidative stress generated during the biotransformation of azo dyes into reactive intermediates. These intermediates promote the generation of reactive oxygen species (ROS), leading to lipid peroxidation and oxidative damage to cellular macromolecules. The significant increase in malondialdehyde (MDA) levels, along with decreased activities of antioxidant enzymes such as superoxide dismutase (SOD), catalase and reduced glutathione (GSH), further substantiates the presence of oxidative imbalance in tartrazine-treated rats. Similar findings have been reported by Chequer *et al.* (2011), highlighting the role of azo dye metabolites in inducing oxidative stress and cellular toxicity. In contrast, treatment with *Camellia sinensis* markedly attenuated the adverse effects of tartrazine, as evidenced by normalization of liver enzyme levels, restoration of antioxidant status, and improvement in liver histological architecture. These hepatoprotective effects are attributed to the presence of polyphenolic compounds, particularly epigallocatechin gallate (EGCG), which exhibit potent antioxidant properties. EGCG exerts its protective action through free radical scavenging, inhibition of lipid peroxidation, and enhancement of endogenous antioxidant defense mechanisms. These findings are in agreement with Abdel-Raheem, Abdel-Ghany and Mohamed (2010), who demonstrated reduced oxidative stress and tissue protection with green tea extract.

Similarly, Vieira, Teixeira and Ferreira (2011) reported improved liver function and decreased enzyme leakage following green tea treatment. Furthermore, the combined treatment group (tartrazine + green tea) showed partial to complete restoration of biochemical, oxidative, and histopathological parameters, indicating the protective efficacy of natural antioxidants. Overall, the results suggest that *Camellia sinensis* possesses significant therapeutic potential in mitigating tartrazine-induced liver toxicity and may serve as a natural protective agent against food additive-induced hepatotoxicity.

V. NOVEL FINDINGS

This study provides several important contributions. Firstly, it demonstrates time-dependent toxicity of tartrazine over multiple exposure durations

(7–60 days), offering a comprehensive understanding of progressive liver damage. Secondly, it integrates biochemical and oxidative stress analysis, providing a multidimensional assessment of toxicity. Finally, the study clearly establishes a hepatoprotective mechanism of green tea, emphasizing its role in restoring antioxidant balance and preventing cellular damage.

VI. CONCLUSION

The findings of the present study indicate that tartrazine, a commonly used synthetic food dye, can induce significant liver toxicity in albino rats. This toxicity is evident from the elevated levels of liver enzymes such as ALT, AST and ALP, along with noticeable alterations in liver histology, including cellular degeneration, necrosis, and inflammatory changes. These effects are mainly associated with increased oxidative stress and an imbalance in the antioxidant defense system.

However, treatment with *Camellia sinensis* (green tea) showed a considerable protective effect against tartrazine-induced liver damage. Improvement in biochemical parameters and restoration of liver structure suggest that green tea possesses strong hepatoprotective properties. This effect is likely due to the presence of bioactive compounds such as catechins and epigallocatechin gallate (EGCG), which act as powerful antioxidants.

In conclusion, the study highlights the harmful impact of excessive consumption of synthetic food additives and emphasizes the beneficial role of natural antioxidants in reducing liver damage. Further studies are recommended to explore the detailed mechanisms involved and to assess the long-term safety and potential clinical applications of such natural compounds.

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