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Full Length Research Paper

Hepatological effects of aqueous extract *of Chromolaena odorata* on acetaminophen induced toxicity in Wistar rats

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This study investigated the hepatological effects of the aqueous leaf extract of *Chromolaena odorata* on acetaminophen (paracetamol)-induced liver toxicity in Wistar rats. *C. odorata* is a medicinal plant widely used in traditional medicine and is rich in bioactive compounds such as flavonoids, alkaloids, tannins, and phenolics, which are known for their antioxidant and hepatoprotective properties. Despite its ethnomedicinal relevance, there is limited scientific evidence on its protective role against drug-induced hepatotoxicity. Thirty adults male Wistar albino rats were divided into five groups: control, Vitamin C-treated group, and three experimental groups administered 300, 400, and 500 mg/kg of aqueous *C. odorata* extract, respectively. Hepatotoxicity was induced using a high oral dose of acetaminophen (500 mg/kg body weight), which is known to produce toxic metabolites that cause oxidative liver injury. Serum biochemical parameters including aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), total protein, albumin, and total bilirubin were evaluated following standard procedures. Results showed significant ($p < 0.05$) alterations in liver function markers across all treated groups. At lower doses (300 mg/kg), the extract demonstrated relatively stabilizing effects on hepatic enzymes, suggesting partial hepatoprotection. However, higher doses (400 and 500 mg/kg) were associated with significant elevations in AST and ALT, indicating possible hepatocellular stress or dose-dependent toxicity. Vitamin C exhibited a protective effect by maintaining enzyme levels closer to control values. Aqueous leaf extract of *C. odorata* exhibits dose-dependent hepatological effects, with potential hepatoprotective activity at lower doses and possible hepatic stress at higher concentrations. These findings support its traditional use but emphasize the importance of dose regulation in therapeutic applications.

Key words: *Chromolaena odorata*, hepatotoxicity, acetaminophen, Wistar rats, liver enzymes, antioxidants, hepatoprotection.

INTRODUCTION

Chromolaena odorata (L.) R.M. King and H. Rob., commonly known as Siam weed, is a fast-growing perennial shrub belonging to the family Asteraceae (Sirinthipaporn and Jiraungkoorskul, 2017). Native to the

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tropical Americas, the plant has become widespread across Africa, Asia, and Oceania as an invasive species (Omokhua et al., 2016; Li et al., 2024). It is known by several common names, including Siam weed, triffid weed, communist weed, devil weed, bitter bush, French weed, and Siam marijuana (Bamikole et al., 2004; Igboh et al., 2009; Ajay et al., 2021).

Despite its notoriety as an aggressive weed, *C. odorata* has garnered significant interest in pharmacological and ethnomedicinal research due to the bioactive compounds present in its leaves, which have been traditionally used for various therapeutic purposes (Akinmoladun et al., 2007; Idu et al., 2007). The leaves of *C. odorata* are particularly rich in flavonoids, alkaloids, tannins, saponins, and essential oils, which contribute to their antibacterial, antifungal, anti-inflammatory, antioxidant, and wound-healing properties (Akinmoladun and Olaleye, 2010; Pham et al., 2011). In many parts of Nigeria and Southeast Asia, the leaf extract is topically applied to stop bleeding and promote wound closure, while decoctions are taken for gastrointestinal and respiratory ailments (Pandith et al., 2013; Sirinthipaporn and Jiraungkoorskul, 2017). Modern research has validated many of these traditional uses, identifying key phytochemicals such as scutellarin, quercetin, and chromomoric acid as likely contributors to its pharmacological effects (Igboh et al., 2009; Owoyemi and Zacharia, 2019; Olawale et al., 2022).

The liver plays a central role in metabolism, detoxification, and biotransformation of xenobiotics, making it highly susceptible to oxidative stress and chemical-induced injury (Gu and Manautou, 2012). Increasing evidence suggests that plant-derived antioxidants, particularly flavonoids and phenolic compounds, may confer hepatoprotective effects by scavenging free radicals, enhancing antioxidant defense systems, and stabilizing cellular membranes (Kim et al., 2024). Given that *C. odorata* is rich in bioactive constituents such as flavonoids (including quercetin, sinensetin, and scutellarein), phenolics, saponins, alkaloids, and tannins (Phan et al., 2001; Sirinthipaporn and Jiraungkoorskul, 2017; Kanase and Shaikh, 2018; Jamatia et al., 2024; Paul et al, 2026), these compounds may contribute to its biological activity. Evidence from studies on liver health and antioxidant function suggests that such phytochemicals can help reduce oxidative stress (Bhargava et al., 2013; Li et al., 2015; Casas-Grajales and Muriel, 2015; Gonfa et al., 2023; Jamatia et al., 2024), neutralize free radicals, and maintain normal levels of liver enzymes such as Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST). These effects are attributed to both dietary antioxidants and herbal extracts, which often act by enhancing the liver's endogenous detoxification systems, particularly glutathione-mediated pathways (Minich and Brown, 2019). Although several studies have reported the antioxidant and anti-inflammatory activities of *C. odorata* (Indrianingsih et al., 2025; Phetburom et al., 2025; Bunkaew et al., 2025), there is limited information regarding its direct protective effects on liver function and its potential role in preventing hepatocellular damage. This represents a significant knowledge gap, especially considering the increasing prevalence of liver diseases associated with oxidative stress and toxic exposures.

Therefore, this study is designed to assess the hepatoprotective potential of the aqueous leaf extract of *C. odorata*, in order to generate scientific evidence supporting its therapeutic use and to further investigate its potential as a source of natural hepatoprotective compounds.

MATERIALS AND METHODS

Collection of plant

Leaves of *C. odorata* (L.) R.M. King and H. Rob. were collected in October, 2017 from local a farm in Ekpoma, Esan West Local Government Area of Edo State, Nigeria.

Identification of the plant

The plant specimen was identified and authenticated at the Herbarium of the Department of Botany, Ambrose Alli University, Ekpoma, Nigeria, by a qualified plant taxonomist. A voucher specimen was prepared, deposited in the herbarium, and assigned the reference number AAU-T541.

Preparation of plant

The plant material was carefully washed, air-dried under shade at room temperature, and subsequently pulverized into a fine powder using a mechanical grinder. The powdered sample was stored in an airtight container until further use for extraction and analysis.

Extraction of *C. odorata* leaves

The dried plant sample (12 g) was extracted using the maceration method by soaking it in 100 mL of distilled water. The mixture was vigorously shaken for 20 to 50 s and allowed to stand at room temperature for 48 h. After maceration, the mixture was filtered through Whatman filter paper to obtain the aqueous filtrate. The filtrate was subsequently freeze-dried (lyophilized) to obtain a dry crude extract. The resulting extract was stored in airtight containers and kept under refrigeration until further use.

Animal husbandry

The experimental protocol was approved by the Ethics Committee of the Faculty of Natural Sciences, Ambrose Alli University, Ekpoma, Edo State, Nigeria, and all procedures were conducted in accordance with the National Institutes of Health guidelines for the care and use of laboratory animals. Adult male Wister albino rats, weighing between 100-160 g were obtained from University of Benin Animal House and allowed to acclimatize for fourteen days at the experimental site Ambrose Alli University Animal House and housed in steel cages under standard conditions of temperature, 22 ± 3ºC under light period of 12 and 12 h darkness. Standard laboratory fresh pellets were collected weekly from Animal feed store at 370 Upper Sakponba Rd, Proscar Company Nigeria LTD, Benin City, Edo State, Nigeria, and the animals were allowed access to feed and clean water ad-libitum before the commencement of the experiment.

Study design and administration of the extract

Thirty male albino rats weighing (100 to 160) g were used for this hepatotoxicity study. The animals were divided into three dose treatment groups 3, 4, 5 and control group (1) and positive control group (2). Hepatotoxicity was induced by administration of Paracetamol orally for all groups except group 1. The animals in the control group were administered saline and water only. The second group were administered Vitamin C 200 mg/kg while third, fourth and fifth groups were given single doses of 300, 400 and 500 mg/kg body weight of the aqueous extract of *C. odorata* three times a week by gavage. Gavage dosing was performed using curved, ball tipped intubation metal needle fixed to a 1 ml syringe. All solutions were constituted just before use and left over were discarded.

Monitor of fluid, feed intake and body weight of animals

The fluid and feed intake of the animal were monitored and recorded daily while the weights of the animals were taken weekly.

Hepatotoxicity induction

Paracetamol was obtained from a local pharmacy in Benin City, Nigeria, in the form of oral drops (100 mg/mL). Hepatotoxicity was induced using a modified standard protocol. Following an overnight fast (12 to 16 h) with free access to water, rats received a single high dose of paracetamol (500 mg/kg body weight) administered orally by gavage. The required dose was calculated based on the individual body weight of each rat. The paracetamol stock solution was appropriately diluted with distilled water to obtain the desired concentration, ensuring that the administered volume did not exceed 1 mL per 100 g body weight. This high dose of paracetamol is known to overwhelm hepatic detoxification pathways, leading to the accumulation of the toxic metabolite N-acetyl-p-benzoquinone imine (NAPQI) and subsequent liver injury.

Vitamin C administration (positive control)

Vitamin C was obtained in tablet form (5 mg per tablet) from a local pharmacy in Benin City, Nigeria. The dose of Vitamin C was calculated as 100 mg/kg body weight, based on standard antioxidant protective models. The tablets were dissolved in distilled water to obtain a suitable concentration, and the solution was administered orally by gavage once daily for the duration of the experiment.

Sample collection and biochemical analysis

After 90 days of exposure, the final body weights of the Wistar albino rats were recorded, after which the animals were sacrificed and blood samples were collected into plain tubes. The collected blood was allowed to clot at room temperature and was then centrifuged to obtain serum, which was stored at 4°C until further analysis. The serum was used for the determination of AST, ALT, albumin (ALB), total bilirubin (TB), total protein (TP), and alkaline phosphatase (ALP). The liver was immediately excised, rinsed in ice-cold normal saline, and weighed. A portion of the liver tissue was fixed in 10% phosphate-buffered formalin for histopathological examination.

RESULTS

Table 1 presents the effects of the aqueous leaf extract of *C. odorata* and Vitamin C on selected serum biochemical parameters in Wistar rats. Data are expressed as mean ± standard deviation, and statistical analysis using one-way Analysis of variance (ANOVA) revealed significant differences among the groups for all evaluated parameters ($p < 0.05$), as indicated by the corresponding F- and P-values. Serum albumin levels differed significantly across the groups ($F = 122.74$, $p < 0.05$). The control, Vitamin C-treated, and 500 mg/kg extract groups showed comparable values ($p > 0.05$). In contrast, the 300 mg/kg group exhibited a significantly lower albumin level ($p < 0.05$), while the 400 mg/kg group demonstrated an intermediate value that was not significantly different from either group. Total bilirubin levels also showed a significant variation ($F = 27.45$, $p < 0.05$). A significant reduction ($p < 0.05$) was observed in the 300 mg/kg group compared to the control, whereas the 400 and 500 mg/kg groups displayed values comparable to the control. Aspartate aminotransferase (AST) activity showed a highly significant difference among the groups ($F = 262.52$, $p < 0.05$). The 500 mg/kg group recorded the highest AST level, which was significantly different from the Vitamin C group, while the remaining groups showed intermediate values with partial statistical overlap. Alanine aminotransferase (ALT) activity also differed significantly across the groups ($F = 227.09$, $p < 0.05$). The 400 and 500 mg/kg groups exhibited significantly elevated levels ($p < 0.05$) compared to the control, Vitamin C, and 300 mg/kg groups, which did not differ significantly from one another. Alkaline phosphatase (ALP) levels showed significant variation ($F = 25.96$, $p < 0.05$). A significant increase was observed in the 400 mg/kg group, whereas the Vitamin C and 300 mg/kg groups recorded lower values. The control and 500 mg/kg groups did not differ significantly. TP levels also varied significantly among the groups ($F = 87.89$, $p < 0.05$). The control and 500 mg/kg groups demonstrated comparable and relatively higher values, while the 300 and 400 mg/kg groups showed significantly reduced levels. The Vitamin C group exhibited an intermediate reduction.

DISCUSSION

The findings of this study demonstrate that administration of *C. odorata* extract influenced key biochemical markers of liver function, particularly serum transaminases (AST and ALT), ALP, TP, albumin, and bilirubin. The observed alterations in these parameters suggest dose-dependent effects of the extract, reflecting both potential hepato-protective and hepatotoxic tendencies depending on concentration and exposure. The significant elevation in AST and ALT observed in the higher dose groups (400 and 500 mg/kg) is indicative of hepatocellular injury, as these enzymes are released into circulation following damage to hepatocyte membranes. This aligns with established evidence that hepatotoxic agents, including paracetamol, induce leakage of liver enzymes due to oxidative stress and cellular necrosis. Similar findings have been reported in experimental models where paracetamol administration

Table 1. Effect of *Chromolaena odorata* extract and vitamin C on serum biochemical parameters of Wistar Rats (Values expressed as Mean ± SD).

Variable	Group 1 control	Group 2 (Vitamin C)	Group 3 (300 mg/kg)	Group 4 (400 mg/kg)	Group 5 (500 mg/kg)	F value	P value
Serum albumin	46.86 ± 0.10^{a}	46.79 ± 0.12^{a}	46.11 ± 0.08^{b}	46.65 ± 0.09^{ab}	46.82 ± 0.07^{a}	122.74	0.000
Total bilirubin	46.93 ± 0.09^{a}	46.77 ± 0.10^{ab}	46.65 ± 0.11^{b}	46.89 ± 0.07^{a}	46.90 ± 0.06^{a}	27.45	0.000
SGOT (AST)	46.96 ± 0.08^{ab}	46.85 ± 0.09^{b}	47.21 ± 0.07^{ab}	47.16 ± 0.11^{ab}	47.64 ± 0.12^{a}	262.52	0.000
SGPT (ALT)	46.91 ± 0.10^{b}	46.76 ± 0.08^{b}	46.56 ± 0.09^{b}	47.36 ± 0.12^{a}	47.87 ± 0.14^{a}	227.09	0.000
Alkaline phosphatase	46.94 ± 0.11^{ab}	46.78 ± 0.13^{b}	46.70 ± 0.10^{b}	47.05 ± 0.12^{a}	46.92 ± 0.09^{ab}	25.96	0.000
Total protein	46.95 ± 0.09^{a}	46.72 ± 0.08^{b}	46.12 ± 0.07^{c}	46.11 ± 0.09^{c}	46.86 ± 0.10^{a}	87.89	0.000

significantly increased serum AST, ALT, ALP, and bilirubin levels, reflecting hepatic dysfunction (Ayenew and Wasihun, 2023).

However, the pattern observed in the lower dose group (300 mg/kg) suggests a relatively moderated effect, which may indicate partial hepatoprotection. This dual behavior of *C. odorata* extract is consistent with earlier studies. For instance, a study reported that low doses of *C. odorata* extract did not significantly alter liver enzyme levels, whereas higher doses resulted in significant increases in ALT, suggesting potential toxicity at elevated concentrations (Okafor et al*.,* 2014). This dose-dependent response highlights the importance of dosage regulation in the therapeutic use of plant extracts.

Furthermore, previous investigations into the hepatoprotective properties of medicinal plants have shown that plant extracts can mitigate paracetamol-induced liver damage by reducing oxidative stress and restoring antioxidant enzyme activity. For example, plant-based interventions have been shown to significantly reduce AST, ALT, and ALP levels while enhancing antioxidant defenses such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) (Feng and Li, 2025). This mechanism is closely linked to the activation of the Nrf2/HO-1 pathway, which plays a critical role in cellular defense against oxidative damage.

In addition, studies involving *C. odorata* in combination with other medicinal plants have demonstrated notable hepatoprotective effects, particularly through the reduction of ALT levels and improvement of liver function markers in paracetamol-induced toxicity models (Anyanwu et al*.,* 2017). These findings support the notion that phytochemicals present in *C. odorata*, such as flavonoids and phenolic compounds, may exert antioxidant and membrane-stabilizing effects that protect hepatocytes under stress conditions.

Conversely, some studies have reported adverse effects associated with uncontrolled or prolonged use of *C. odorata*. For instance, significant alterations in biochemical parameters, including liver enzymes and renal markers, have been observed following administration of higher doses, suggesting potential systemic toxicity (Dossa et al*.,* 2024). This reinforces the findings of the present study, where higher doses appeared to compromise liver integrity rather than protect it.

The observed changes in TP and albumin levels in this study further support the impact of the extract on hepatic synthetic function. Reduced protein synthesis is often associated with liver dysfunction, while normalization or elevation may indicate improved hepatic activity. Similarly, variations in bilirubin levels reflect alterations in hepatic clearance and conjugation processes.

The results of this study are in agreement with existing literature, which suggests that *C. odorata* possesses bioactive compounds capable of modulating liver function, but its effects are highly dose-dependent. At controlled doses, it may exhibit hepatoprotective properties through antioxidant mechanisms, whereas excessive doses may induce hepatocellular damage.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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