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Ameliorative Effect of Ginger and Turmeric on Serum C-Reactive Protein, Total Protein and Albumin in Lead Induced Wistar Rats.

Awalu, Chimezie Joseph[1] and Ekechi, Blessing[2]
[1] Faculty of Medical Laboratory Science, Chemical Pathology Department, Abia State University, Abia State, Nigeria.
[2]Medical Laboratory Science Department, Ebonyi State University, Abakaliki, Nigeria.

Corresponding author: Awalu Chimezie Joseph, Faculty of Medical Laboratory Science, Chemical Pathology Department, Abia State University, Abia State, Nigeria

ABSTRACT: Lead toxicity, induces oxidative stress and systemic inflammation, resulting in altered serum protein profiles and elevated inflammatory biomarkers. Ginger and turmeric are widely used natural antioxidants with known anti-inflammatory effect. This experimental study evaluated the ameliorative effects of ginger (Zingiber officinale) and turmeric (Curcuma longa) on serum C-reactive protein, total protein and albumin in lead-induced wistar rats. Twenty-five rats were randomized into five groups (n=5): Group 1 (control), Group 2 (Lead only), Group 3 (lead + ginger), Group 4 (Lead + turmeric), Group 5 (Lead + ginger + turmeric). Lead acetate (30mg/kg body weight) was administered orally for 14 days; treatments with ethanolic extracts of ginger ad turmeric (200mg/kg body weight) were given concurrently where applicable. Serum CRP was measured by ELISA; total protein and albumin were determined by Biuret and bromocresol green methods respectively. Data were analyzed using IBM SPSS version 25. One-way Analysis of Variance (ANOVA) was used to determine the significance of difference in the mean of all parameters. Lead exposure produced significant elevations in CRP and reductions in total protein and albumin ($p<0{\cdot}05$). Treatment with ginger and turmeric individually improved these biomarkers, while combined treatment restored values close to control. These findings indicate that ginger and turmeric have anti-inflammatory and hepatoprotective effects against lead-induced biochemical alterations.

Keywords: Lead toxicity, Ginger, Turmeric, Ameliorative effects, C reactive protein

INTRODUCTION

Antioxidants are molecules that inhibit the oxidation of other molecules, thereby protecting the cells from damage caused by free radicals and reactive oxygen species (Aljutaily, 2022). Antioxidants, including essential oils, polyphenols, and vitamins, inhibit the oxidation of molecules in cells. By limiting radical or molecule formation, antioxidants reduce oxidative stress and prevent tissue damage; they have potential for improving immune function when acting as immunostimulants (Aljutaily, 2022). Turmeric and Ginger, two widely used culinary and medicinal rhizomes, contains phytochemicals such as curcumin, gingerols and shogaols, which are known for their free radical scavenging, enzyme-modulating, and anti-inflammatory effects (Ajanaku *et al*., 2022; Ballester *et al*., 2023). These compounds have been shown to enhance antioxidant enzyme activity, reduce lipid peroxidation and mitigate tissue damage in chemically induced toxicity models. Ginger (*Zingiber officinale Roscoe*) serves as both a spice, food, and traditional medicine globally, possessing anti-oxidant, antifungal, antibacterial and anti-inflammatory properties (Soltanian *et al*., 2019). Bioactive compounds in ginger include; gingerol, gingerdiol, zingiberene, shogaol, zingerone and gingerols. Many fatty acids and amino acids also contribute to gingers antioxidant activity (O'Rotimi *et al*., 2015). Turmeric (Curcuma longa) in its active form "Curcumin", has been shown to modulate oxidative pathways and restore redox balance, while gingerol in ginger exhibits strong radical-scavenging activity and supports cellular antioxidant responses (Ayustaningwarno *et al*., 2024).

Lead is a multi-organ toxicant involved in various cancers, neuronal and renal damages and reproductive impairments in both human and animals, which can eventually cause death in young children (Shaffer *et al*., 2017). Once lead is absorbed, it moves into the bloodstream where it is primarily circulated to the soft tissues through the plasma (kidney, liver and brain), blood and mineralized tissues such as the bones. Then its exerts more toxic effects by binding to the cell membrane, damaging protein architectural structure and meddling with gene interpretation in the body (Ekanem *et al*., 2015). In Nigeria, developing states like Ebonyi state, is well known for its multiple mining sites such as; Enyigba, Mkpuma Akpatakpa and Ameka for lead and zinc. Lead contamination in Ebonyi state is a serious health concern, particularly in areas like Enyigba and Ameka, where studies found high levels in soil, water, plants and human blood (Enyankware and Obasi, 2022). Contamination occurs through ingestion of lead-contaminated water and food grown in polluted soil with significant contributions from surface water, plants and groundwater to human exposure.

Ebonyi state lacks social amenities such as, good pipe borne water (well-known for hard water), good drainage systems and most of the lead contaminated water flows to homes and is ingested by humans. Also, these lead infected soils are used to grow food items which are transported to the markets for sale. Most times, the waste products from these mining sites are deposited in rivers, of which people living in rural communities use for cooking, drinking and washing. The aim of this study is to determine the serum levels of C-reactive

protein, total protein, and albumin in lead-induced Wistar rats and to compare the individual and combined ameliorative effects of ginger (*Zingiber officinale*) and turmeric (*Curcuma longa*) on lead-induced toxicity.

MATERIALS AND METHODS

Materials

The Lead acetate and concentrated methanol were purchased from Asungun Enterprise Ltd, the ginger and turmeric were purchased from International Market in Abakaliki. The ginger (*Zingiber officinale Roscoe*) and turmeric (*Curcuma longa*) rhizomes were taken to a taxonomist for identification and lead acetate and methanol were taken to an Industrial chemist for confirmation before preparation of the extract and experiment began. Twenty-five (25) healthy Wistar rats weighing 100-120g, were procured from Department of Veterinary Medicine, University of Nigeria Nsukka. The rats were housed at Animal Farm Presco Campus, Ebonyi state university in a steel cage. The average temperature recorded in the room was 28^0 C and 12h light-dark cycle with access to water and free pellet food diets with average humidity 40-60% was recorded during the experiment. They were left for two weeks for acclimatization before starting the experiment. Only apparently healthy, non-pregnant and mature Albino Wistar rats without any known deformities, weighing 100-120g were used for this study

Preparation of Lead acetate, Ginger and Turmeric Extracts

10g of lead acetate was weighed on a weighing balance and was dissolved in 100ml of distilled water.

Fresh rhizomes of ginger (*Zingiber officinale*) and turmeric (*Curcuma longa*) were washed thoroughly under running tap water to remove dirt, peeled, sliced into small pieces, and dried at room temperature (18-30°C) for two weeks. The dried samples were milled into coarse powders using a commercial electric blender. Thereafter, 100 g of each powdered sample was separately macerated in 200ml of concentrated methanol and placed on a water bath at 65°C for 24 hours to obtain the extracts. The resulting methanolic extracts were preserved in a refrigerator at 4°C prior to administration.

Experimental Protocol

Based on the oral LD_{50} of lead acetate which was 600 mg/kg body weight for Wistar rats (Umar *et al*., 2019), 5% (30mg/kg) of the LD_{50} was used in our study. The dose for ginger and turmeric used was 200mg/kg body weight (Ahmad *et al*., 2024). Twenty-five (25) Wistar rats were randomly grouped into five groups and treated as described in table 1.0 below.

Table 1.0: Study design protocol

Blood

Groups	Names	Description
Group 1	Positive Control	Received water and feed only for 14 days consecutively.
Group 2	Negative control	Received lead (30mg/kg body weight) and feed only for 14 days consecutively.
Group 3	Treatment 1	Received lead (30mg/kg body weight) and ginger extract (200mg/kg body weight) for 14 days consecutively.
Group 4	Treatment 2	Received lead (30mg/kg body weight) and turmeric extract (200mg/kg body weight) for 14 days consecutively.
Group 5	Treatment 3	Received lead (30mg/kg body weight), turmeric and ginger (200mg/kg body weight) extract for 14 days consecutively.

Sample collection

At the end of the fourteenth day, the Wister rats were bled by retro-orbital puncture under isoflurane anesthesia and carbon dioxide euthanasia chamber. Blood samples were collected into a plain laboratory container, allowed to clot, retract and centrifuged at 4000rpm for 10 minutes to separate the serum from the red blood cells. The sera were harvested into small aliquots plain container and stored at -20ºC prior to analysis.

Laboratory Analysis

C-reactive Protein (CRP) was estimated using Enzyme Linked Immunosorbent Assay (ELISA) as described by (Gewurz *et al*., 1982), Total protein (TP) and Albumin (Alb) were measured using a spectrophotometric method as described by Doumas *et al*., 1981 and Doumas *et al*., 1971 respectively

Result

One-way analysis of variance results of serum CRP, total protein, and albumin across the experimental groups are presented in Table 1.2. The result revealed a statistically significant difference in mean serum C-reactive protein (CRP) levels across the five experimental groups ($p < 0.001$). The negative control group (Group 1) showed a mean CRP value of 0.21 ± 0.02 mg/L, while the positive control group exposed to lead only (Group 2) showed a significantly higher mean CRP concentration (0.31 ± 0.03 mg/L, $p < 0.05$). Rats treated with lead plus ginger extract (Group 3) exhibited a further elevation in CRP levels (0.55 ± 0.04 mg/L), which differed significantly from both control groups ($p < 0.05$). Similarly, the turmeric-treated group (Group 4) showed a significantly increased CRP

concentration (0.76 ± 0.03 mg/L) compared with Groups 1 and 2 ($p < 0.05$). The combined ginger and turmeric treatment group (Group 5) revealed a mean CRP value of 0.17 ± 0.02 mg/L, which was not significantly different from the negative control ($p > 0.05$) but differed significantly from the positive control and other treatment groups ($p < 0.05$).

In serum total protein levels, a statistically significant difference was observed among the experimental groups ($p < 0.001$). The negative control group showed a mean total protein value of 63.63 ± 1.33 g/L, while the positive control group showed a significantly reduced mean value (57.13 ± 1.63 g/L, $p < 0.05$). Treatment with ginger extract (Group 3) resulted in a further decrease in total protein concentration (51.53 ± 1.55 g/L), which was significantly lower than both control groups ($p < 0.05$). The turmeric-treated group (Group 4) also demonstrated a reduced mean total protein level (54.50 ± 1.29 g/L) compared with the negative control ($p < 0.05$). In contrast, animals treated with the combined ginger and turmeric extract (Group 5) showed a significantly higher total protein concentration (68.40 ± 2.07 g/L) compared with the positive control and other treatment groups ($p < 0.05$), and was comparable to the negative control ($p > 0.05$).

Serum albumin levels similarly showed a significant difference across the groups ($p < 0.001$). The mean albumin concentration in the negative control group was 46.33 ± 1.53 g/L, whereas the positive control group exhibited a marked reduction (34.25 ± 0.96 g/L, $p < 0.05$). Rats administered lead with ginger extract (Group 3) showed a further reduction in albumin levels (29.78 ± 1.00 g/L), which differed significantly from both controls ($p < 0.05$). The turmeric-treated group (Group 4) had a mean albumin value of 33.75 ± 0.96 g/L, which was significantly lower than the negative control but comparable to the positive control ($p > 0.05$). The combined treatment group (Group 5) demonstrated a significant increase in albumin concentration (43.40 ± 1.52 g/L) relative to the positive control and other treatment groups ($p < 0.05$), with values approaching those of the negative control.

Table 1.2: C-Reactive Protein, Total Protein, and Albumin Levels Across Experimental Groups

Parameter	Group 1	Group 2	Group 3	Group 4	Group 5	p-value
CRP (mg/L)	0.21 ± 0.02^{a}	0.31 ± 0.03^{b}	0.55 ± 0.04^{c}	0.76 ± 0.03^{d}	0.17 ± 0.02^{a}	< 0.001*
Total Protein (g/L)	63.63 ± 1.33^{b}	57.13 ± 1.63^{a}	51.53 ± 1.55^{a}	54.50 ± 1.29^{a}	68.40 ± 2.07^{c}	< 0.001*
Albumin (g/L)	46.33 ± 1.53^{c}	34.25 ± 0.96^{a}	29.78 ± 1.00^{a}	33.75 ± 0.96^{a}	43.40 ± 1.52^{b}	< 0.001*

Values are presented as mean ± standard deviation (SD). *Mean difference significant at $p < 0.05$. Within each row, means with the same superscript are not significantly different, while means with different superscripts differ significantly at $p < 0.05$ (Tukey HSD). Group 1 (negative control): feed and water only; Group 2 (positive control): lead (30 mg/kg body weight); Group 3: lead (30 mg/kg body weight) and ginger extract (200 mg/kg body weight); Group 4: lead (30 mg/kg body weight) and turmeric extract (200 mg/kg body weight); Group 5: lead (30 mg/kg body weight) and combination of ginger and turmeric extracts (200 mg/kg body weight).

Discussion

The control group exhibited normal protein parameters, characterized by high total protein and albumin concentrations alongside low serum C-reactive protein (CRP). These findings indicate intact hepatic biosynthetic function and the absence of inflammatory stress. In healthy rats, balanced protein synthesis is maintained, with albumin and total protein serving as key indicators of liver synthetic capacity, while CRP reflects acute-phase inflammatory activity. This observation aligns with reports by Abdelhamid *et al*. (2020) and Abubakar *et al*. (2020), who demonstrated that baseline hepatic protein profiles in control animals are marked by robust albumin and total protein levels, reflecting unimpaired liver function.

Lead exposure resulted in a significant elevation of serum CRP, indicating the induction of systemic inflammation. Concurrently, significant reductions in total protein and albumin levels were observed, suggesting hepatic injury and compromised protein synthesis. Lead is known to disrupt hepatic function through mechanisms including oxidative stress, mitochondrial dysfunction, and inhibition of ribosomal RNA transcription, ultimately impairing protein biosynthesis. Okediran *et al*. (2022) demonstrated that lead acetate exposure significantly reduces serum albumin and total protein due to hepatocellular necrosis and altered metabolic enzyme activity. Similarly, Oladipo *et al*. (2016) reported marked hypoproteinemia in lead-treated rats. The elevated CRP levels further confirm inflammatory activation, likely mediated by lead-induced cytokine release, as reported by Kalahasthi *et al*. (2018).

Ginger supplementation altered protein parameters, with reductions in total protein and albumin and an elevated CRP level relative to the control group. Although these values remained deranged, ginger-treated rats showed partial improvement in hepatic synthetic function compared with the lead-only group. Ginger's bioactive compounds, including gingerols and shogaols, possess antioxidant and anti-inflammatory properties that mitigate lead-induced hepatotoxicity by reducing oxidative damage and inflammatory signaling. This is supported by Abdel-Daim *et al*. (2019) and Adeyemi *et al*. (2020), who demonstrated that ginger extract improves serum protein profiles and attenuates hepatic oxidative stress in toxicant-exposed models.

Turmeric (*curcumin*) treatment also produced improved, though still suboptimal, protein parameters. *Curcumin* exerts potent antioxidant and anti-inflammatory effects by suppressing lipid peroxidation and inhibiting pro-inflammatory signaling pathways. In models of lead toxicity, *curcumin* has been shown to restore serum protein concentrations and improve hepatic histological architecture

(Akinrinde *et al*., 2020). The observed reduction in CRP may be attributed to curcumin's ability to downregulate inflammatory cytokines such as IL-6 and TNF-α, which are key regulators of hepatic CRP expression (Rahmani *et al*., 2021).

Notably, co-administration of ginger and turmeric produced the most pronounced protective effects, restoring total protein and albumin levels while reducing CRP concentrations to near-control values. This superior outcome suggests a synergistic interaction between the phytochemicals present in ginger and turmeric. The combined antioxidant and anti-inflammatory actions likely enhanced free-radical scavenging, preserved mitochondrial integrity, and improved hepatic synthetic activity. Previous studies have similarly demonstrated that concurrent administration of curcumin and ginger more effectively restores serum proteins and suppresses inflammatory cytokines compared with single treatments (Abdel-Daim *et al*., 2019: Akinrinde *et al*., 2020).

Conclusion

These findings indicate that while ginger or turmeric alone provides partial protection against lead-induced hepatic dysfunction, their combined administration offers superior hepatoprotective and anti-inflammatory benefits. This effect likely reflects additive or synergistic mechanisms, whereby curcumin's strong anti-inflammatory activity complements ginger's potent antioxidant capacity, thereby more effectively counteracting the multifactorial hepatic damage induced by lead exposure.

Recommendation

Future studies should focus on elucidating the underlying molecular mechanisms, determining optimal dosage regimens, and exploring translational relevance in human lead toxicity and related metabolic disorders.

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