Vol. 6 No. 2, Dec. 2026

FUAM Journal of Pure and Applied Science

Available online at
www.fuamjpas.org.ng

An official Publication of
College of Science
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Makurdi.

FUAM Journal of Pure and Applied Science

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Vol.6 No. 2 Dec. 2026

Evaluation of Bioaccumulation of Cadmium in Wistar Rats and Absorption with *Aloe Barbadensis* Extract

Z.N[1*]. Jangber, P.A[2]. Ekwumemgbo, A.O[2]. Oyewale, Z.N[1]. Garba & S.B[3]. Oladele

[1]Department of Chemistry, Ahmadu Bello University, Zaria, Nigeria

[2]Department of Veterinary Anatomy, Ahmadu Bello University, Zaria, Nigeria

[3]Department of Veterinary Toxicology, Ahmadu Bello University, Zaria, Nigeria

[*]**Correspondence E-mail: pastorzaph@gmail.com**

Received: 10/04/2026 **Accepted**: 07/06/2026 **Published online**: 08/06/2026

Abstract

Toxicity from heavy metals, especially cadmium (Cd), poses serious health risks because of its bioaccumulative nature and ability to induce organ damage and oxidative stress. This study evaluated the absorption and protective properties *Aloe barbadensis* extract against cadmium-induced toxicity in Wistar rats. Three groups of five rats each were created. (n = 5): a control group, a cadmium-exposed group, and a treatment group administered with cadmium solution, followed by *Aloe barbadensis* extract (40 mg/L) for 28 days. One-way analysis of variance (ANOVA) was used to evaluate differences among the groups, followed by Turkey's post-hoc test. Using Fourier transform infrared spectroscopy to identify functional groups present in the extract, atomic absorption spectroscopy was used to quantify cadmium accumulation in selected organs. Analysis of Fourier transform infrared spectroscopy confirmed the presence of carboxylic acids (C=O and C–O stretching) and other functional groups as well as secondary amines and alcohols, which may contribute to metal-binding and antioxidant activity. Results revealed significantly higher cadmium accumulation in the organs of untreated rats (0.08, 0.09, and 0.07 mg/L). However, treatment with *A. barbadensis* extract led to a notable reduction in cadmium levels in the liver, kidneys, lungs, and spleen (0.01–0.02 mg/L). Although the extract did not totally stop lung toxicity, notable protective effects were noted in other organs, while spleen tissues showed relative resilience across all groups. These results imply that *A. barbadensis* extract possesses bioactive compounds capable of reducing cadmium accumulation and mitigating tissue damage, indicating its possibility as a natural medicinal substance against cadmium-induced toxicity *in vivo*.

Keywords: Heavy metal, cadmium, *Aloe barbadensis*, *antioxidant, extract*

Introduction

Heavy metal contamination constitutes a major environmental and public health challenge worldwide due to the persistence, bioaccumulation, and toxicity of metals such as cadmium (Cd), chromium (Cr), and lead (Pb). These metals are released into the environment through industrialization, mining activities, battery manufacturing, electroplating, agricultural practices, fossil fuel combustion, and improper waste disposal. Unlike organic pollutants, heavy metals are non-biodegradable and can persist in soil, water, and biological systems for prolonged periods, thereby posing serious ecological and toxicological risks [1–3].

Among these metals, cadmium is regarded as one of the most toxic environmental pollutants because of its high mobility, long biological half-life, and ability to accumulate in vital organs such as the liver, kidneys, lungs, and spleen. Human and animal exposure to cadmium occurs primarily through contaminated food, drinking water, cigarette smoke, industrial emissions, and occupational exposure. Chronic cadmium intoxication has been associated with nephrotoxicity, hepatotoxicity, pulmonary dysfunction, skeletal damage, reproductive abnormalities, immune suppression, and carcinogenicity [4–6]. Cadmium toxicity is largely mediated through the generation of reactive oxygen species (ROS), disruption of antioxidant defense systems, lipid peroxidation, mitochondrial dysfunction, and induction of inflammatory responses, all of which contribute to oxidative stress-induced cellular and tissue injury [7, 8].

Several conventional techniques have been developed for the removal or reduction of heavy metal contamination, including chemical precipitation, membrane filtration, ion exchange, reverse osmosis, adsorption, and electrochemical treatment. Although these methods may be effective under certain

conditions, they are often associated with high operational costs, technical complexity, incomplete metal removal, generation of toxic sludge, and secondary environmental pollution [9, 10]. Consequently, there has been increasing interest in the exploration of natural and environmentally friendly alternatives capable of reducing heavy metal toxicity and enhancing detoxification processes.

Medicinal plants have attracted considerable scientific attention because of their rich phytochemical composition and therapeutic potential. Plant-derived bioactive compounds such as flavonoids, phenolics, tannins, saponins, alkaloids, polysaccharides, and terpenoids possess antioxidant, anti-inflammatory, metal-chelating, and free radical scavenging properties that may help mitigate heavy metal-induced toxicity [11, 12]. These phytochemicals may reduce oxidative damage by neutralizing reactive oxygen species, enhancing endogenous antioxidant enzymes, stabilizing cellular membranes, and binding toxic metal ions, thereby reducing their bioavailability and accumulation in tissues.

Aloe barbadensis (Aloe vera) is one of the most widely utilized medicinal plants globally and has long been recognized for its pharmacological and therapeutic properties. The plant contains a wide variety of biologically active constituents, including polysaccharides, anthraquinones, vitamins, enzymes, amino acids, minerals, flavonoids, and phenolic compounds [13–15]. The gel portion of Aloe vera is particularly rich in polysaccharides such as acemannan, which contribute significantly to its antioxidant, anti-inflammatory, antimicrobial, wound-healing, and cytoprotective activities. Previous studies have demonstrated that Aloe vera extracts can attenuate oxidative stress, reduce lipid peroxidation, improve antioxidant enzyme activities, and protect tissues against chemically induced toxicities [16, 17]. These biological properties suggest that Aloe vera may possess significant potential in reducing cadmium-induced organ damage and oxidative stress.

Despite increasing reports on the phytochemical composition and antioxidant activities of Aloe vera, limited information exists regarding its in vivo protective efficacy against cadmium-induced toxicity, particularly in experimental animal models. Furthermore, the mechanisms through which Aloe vera may influence cadmium absorption, tissue distribution, and toxicological outcomes remain insufficiently understood. Therefore, this study was designed to evaluate the absorption and protective effects of *A. barbadensis* extract against cadmium-induced toxicity in Wistar rats. Specifically, the study aims to assess its potential role in reducing cadmium accumulation and ameliorating toxic effects through its antioxidant and cytoprotective properties. The findings from this study may contribute to the development of safer, cost-effective, and plant-based therapeutic approaches for mitigating heavy metal toxicity.

Materials and Methods

Reagents

All chemicals and reagents used were of analytical grades from JHD Chemicals; this included cadmium (II) chloride, potassium dichromate, lead (II) nitrate, sulfuric acid, disodium hydrogen phosphate, sodium hydrogen phosphate, perchloric acid, chloroform, xylene, ethanol, formalin, sodium hydroxide, hydrochloric acid, nitric acid, distilled water, hematoxylin stain, eosin stain, etc. this text and the other manuscript is the same

Collection, drying, and preparation of plant materials and ethical clearance

Fresh leaves of *Aloe barbadensis* were collected from Sabongari, Zaria, and authenticated at the Herbarium of Ahmadu Bello University (voucher No. ABU02890). Application for ethical approval was made to the institutional committee for animal use and care. It was approved and the voucher number was given (ABUCAUC/2024/049). The leaves were washed, sliced, air-dried, ground into coarse powder, sieved, oven-dried at 80°C for 24 hours, and stored in a polyethylene container [10].

Extraction of plant material/FT-IR analysis of Aloe barbadensis

The powdered plant material was extracted with methanol (1:5 w/v) using cold maceration for 48 hours with periodic shaking. The mixture was filtered, and the filtrate was concentrated using a rotary evaporator, dried to constant weight, and stored in a refrigerator until use [10]. FT-IR analysis of the extract was performed using KBr pellets within the range of 4000–400 cm^{-1} to identify functional groups.

Preparation of stock solutions of cadmium chloride and Aloe barbadensis extract.

A stock solution of cadmium chloride was prepared by dissolving 7.8 mg in 100 cm^3 of distilled water. The *Aloe barbadensis* extract solution was prepared by dissolving 200 mg of extract in dimethyl sulfoxide and diluting to 100 ml with distilled water.

Experimental Animals/Grouping

Fifteen adults female Wistar rats (150–300 g) were obtained from the animal house of the Department of Veterinary Pharmacology and Toxicology, Ahmadu Bello University, Zaria. The rats were acclimatized for two weeks and maintained with feed and water provided *ad libitum*.

The rats were randomly divided into three groups of five animals (n = 5) each and treated orally for 28 days. Group I (positive control) received normal feed and water only. Group II (negative control) was administered 40.0 mg/L cadmium chloride once weekly, with feed and water. Group III (treatment group) received 40.0 mg/L cadmium chloride once weekly and was treated daily with 40.0 mg/L *A. barbadensis* extract, along with feed and water *ad libitum*.

Administration of heavy metal solution and the plant extract solution

The volume administered orally for both the heavy metal solutions and the plant extract solutions was calculated from the equation below [12].

$$\text{Rat dosage } (cm^3) = \frac{Weight\ of\ rat\ (kg) \times drug\ dose}{1kg} \quad (1)$$

Animal sacrifice and harvesting of tissues

After the administration of the heavy metal solution and the absorption with the biomaterial extract, cervical decapitation was used to sacrifice the animals after exposing them to chloroform vapor [13-14]. The rats were dissected using a scalpel blade, and the liver, kidney, spleen, and lungs were harvested [14].

Digestion of tissues

Each tissue sample (2.50 g wet mass) was digested using a mixture of nitric, sulfuric, and perchloric acids (3:1:1). The mixture was heated on a hot plate for 10 minutes until nitric acid evaporated and perchloric acid fumes appeared. After cooling, 25 cm^3 of distilled water was added, and the solution was filtered. The filtrate was then transferred into a 50 cm^3 measuring cylinder and diluted to the mark with distilled water [15-16].

Determination of the levels of cadmium, chromium, and lead in digested tissues

The digested tissues were taken to the Multiuser Science Research Laboratory for heavy metals analysis. Atomic Absorption Spectrometry (AAS) was performed using standard solutions of cadmium, chromium, and lead. Different concentrations were prepared by serial dilution and used to construct calibration curves. The calibration curves showed good linearity with correlation coefficients (R^2) greater than 0.99 [15-17].

Pathological Studies

Portions of the liver, kidney, lungs, and spleen were used for pathological examination. The tissues were fixed in 10% neutral buffered formalin, processed, sectioned with a microtome, and stained with hematoxylin and eosin. The sections were examined under a high-powered microscope, and lesions were photographed using an Amscope.

Results and Discussion

Fourier Transform Infrared (FTIR) Results of Aloe barbadensis Extract

The FT-IR spectrum of Aloe barbadensis extract showed key absorption bands at 1058.6, 1707.1, 2922.2, and 3373.2 cm^{-1}, indicating functional groups involved in metal interaction. The peak at 1707.1 cm^{-1} corresponds to C=O stretching, characteristic of carbonyl groups such as carboxylic acids, aldehydes, ketones, or esters. [18]. Carboxylic acid groups enhance metal binding by donating electron pairs to form stable complexes with metal ions. The broad peak at 3373.2 cm^{-1} indicates O–H stretching of hydroxyl groups and possible N–H stretching of amines, suggesting hydrogen bonding. These hydroxyl groups contribute to metal ion binding through hydrogen bonding and electrostatic interactions, increasing affinity for metal cations [19]. The band at 1058.6 cm^{-1} indicates C–O stretching from alcohols, phenols, or ethers, which provide electron-rich sites for metal ion chelation. The peak at 2922.2 cm^{-1} corresponds to aliphatic C–H stretching, showing organic constituents in the extract. Overall, the presence of C=O, O–H, and C–O functional groups confirms that Aloe barbadensis extract is rich in oxygenated compounds such as carboxylic acids and alcohols, which enhance metal ion binding, chelation, and adsorption, supporting its potential use in heavy metal detoxification.

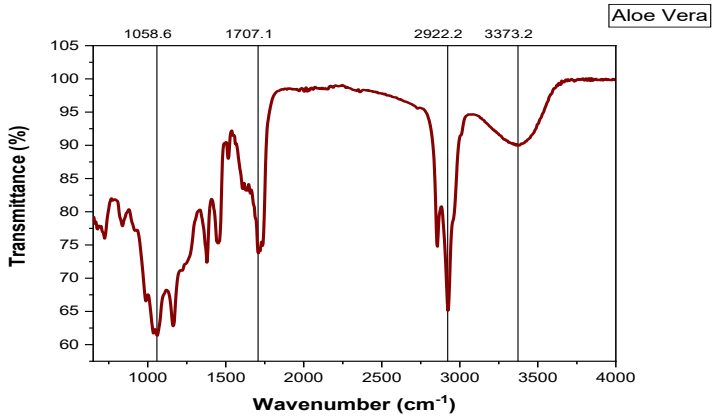

Figure 1: Fourier transform infrared spectrum of *Aloe barbadensis* extract

The effects of in vivo absorption with Aloe barbadensis extract on the concentration of cadmium in the liver, kidney, lungs, and spleen of Wistar rats

The effects of *Aloe barbadensis* extract on cadmium concentration in various tissues are shown in Table 1. The results showed a marked accumulation of cadmium in all

tissues of the negative control group compared to the positive control group. Specifically, cadmium concentrations in the negative control group were 0.090 ± 0.001 mg/L, 0.080 ± 0.001 mg/L, 0.080 ± 0.001 mg/L, and 0.070 ± 0.001 mg/L, whereas the positive control group showed minimal concentrations (0.001 ± 0.0001 mg/L, 0.002 ± 0.0001 mg/L, 0.001 ± 0.0001 mg/L, and 0.002 ± 0.0001 mg/L). This significant elevation confirms the successful induction of cadmium toxicity.

Treatment with *A. barbadensis* extract resulted in a pronounced reduction in cadmium levels across all examined tissues. In the liver, cadmium concentration decreased significantly to 0.020 ± 0.001 mg/L and 0.010 ± 0.001 mg/L in contrast to the negative control (0.090 ± 0.001 mg/L). This reduction suggests an enhanced cadmium clearance effect, likely mediated by phytochemicals such as flavonoids, phenolics, and polysaccharides, which are known to chelate metal ions and promote their mobilization and excretion [20-21].

A similar trend was observed in the kidneys, where cadmium levels in treated rats (0.020 ± 0.001 mg/L) were markedly lower than in the negative control group (0.080 ± 0.001 mg/L). Given that cadmium preferentially accumulates in renal tissues due to its long biological half-life and binding to metallothionein proteins [22], this reduction indicates that *Aloe barbadensis* extract may effectively limit renal cadmium accumulation through metal-binding and detoxification mechanisms.

In the lungs, cadmium concentration was also reduced in the treated group (0.020 ± 0.001 mg/L), suggesting that the extract may enhance systemic cadmium mobilization and reduce tissue deposition. Cadmium-induced pulmonary toxicity is often connected to oxidative stress and inflammation; therefore, the observed reduction may possibly be due to the antioxidant activity of bioactive components like flavonoids and phenolic compounds, which have the ability to bind metal ions and neutralize reactive oxygen species [23-24]. Similar cadmium-lowering effects of *Aloe barbadensis* have been reported by [25], who associated these effects with enhanced metallothionein activity and scavenging free radicals.

Furthermore, cadmium concentration in the spleen was considerably decreased in treated rats (0.030 ± 0.001 mg/L) in contrast to the negative control group (0.070 ± 0.001 mg/L). The spleen, being a key organ involved in immune function and blood filtration, is particularly susceptible to cadmium-induced oxidative damage and immune dysfunction. The observed decrease suggests that *Aloe barbadensis* extract may enhance cadmium clearance and protect splenic tissue, possibly through the action of its antioxidant phytochemicals, which reduce metal bioavailability and oxidative stress [26-27].

The consistent reduction in cadmium levels across all examined organs indicates that *Aloe barbadensis* extract possesses significant metal-chelating and protective properties, supporting its potential role in mitigating cadmium-induced toxicity *in vivo*.

Table 1: The effects of *in vivo* biosorption with *Aloe barbadensis* extract on the cadmium levels in Wistar rats (mg/L)

Groups	Liver	Kidney	Lungs	Spleen
Positive control	0.001±0.01	0.002±0.001	0.003±0.001	0.002±0.001
Negative control	0.08±0.01	0.08±0.001	0.08±0.001	0.07±0.001
A. barbadensis	0.02±0.001	0.02±0.001	0.02±0.001	0.03±0.001

Histopathology of organs of Wistar rats intoxicated with cadmium and treated with Aloe barbadensis extract

The phytomicrographs of the Wistar rats intoxicated with cadmium chloride and treated with *A. barbadensis* extract are presented in Plates 1-3. The findings indicated that there were no obvious lesions in the organs of the positive control rats (Plate 1). This is true of all organs in the absence of toxic agents: preserving their regular physiological processes [28]. The phytomicrographs of the negative control rats (Plate 2) showed congestion in the liver, necrotic tubules in the kidney, thickened interalveolar walls in the lungs, and normal tissue architecture in the spleen. The liver, kidney, and lungs are particularly sensitive to cadmium; toxic effects may be because of their capacity to synthesize metallothioneins, which bind cadmium ions [29, 30]. The hepatic congestion, renal tubular necrosis, and alveolar wall thickening observed corroborate earlier reports by [31, 32,] who observed that cadmium caused congestion and inflammatory cells, tubular necrosis and epithelial degeneration, thickening of alveolar septa, and interstitial inflammation in rats. In the treated group (Plate 3), there was normal tissue architecture in the liver, kidney, and spleen. This may be possibly due to the protective effects of *A. barbadensis* extract against heavy metals [33]. This is in line with [34] and [35], who discovered that there was a restoration of liver and kidney architecture after the administration of *A. barbadensis.* There were thickened interalveolar walls in the lungs; this may suggest ongoing inflammation or fibrosis, which is a known outcome of cadmium-induced pulmonary toxicity. Cadmium can activate alveolar macrophages and epithelial cells, leading to the release of pro-inflammatory cytokines [36]. This agrees with the work of [37], who reported alveolar wall thickening in cadmium-treated animals. Though *A. barbadensis* demonstrated promising therapeutic potential in mitigating cadmium-induced damage in several organs, its efficacy in the lungs may be limited. This is in line with [38], who demonstrated that while plant antioxidants improved liver and kidney histology, lung tissue showed delayed or

incomplete recovery, especially with short treatment duration.

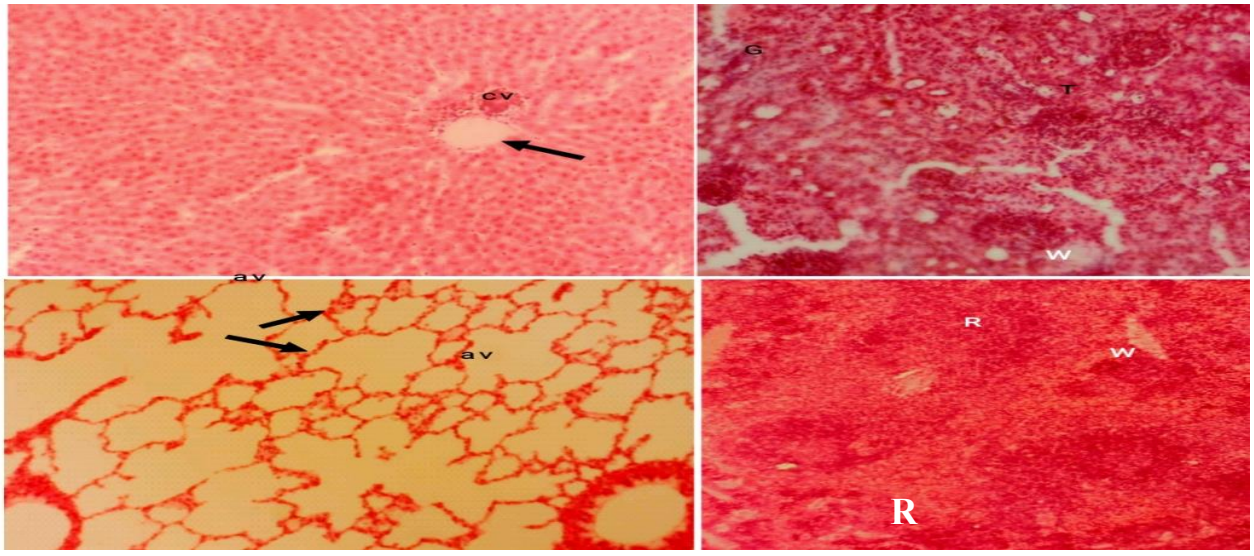

Plate 1: Photomicrographs of liver (A), kidney (B), lung (C), and spleen (D) from the positive control showing normal tissue architectures. Note the central vein (CV) and hepatocytes (arrows) in A (arrow), glomeruli (G), and tubules (T) in B; alveoli (av) and interalveolar walls (arrows) in C; and white (W) and red (R) pulps in D. H & E × 100

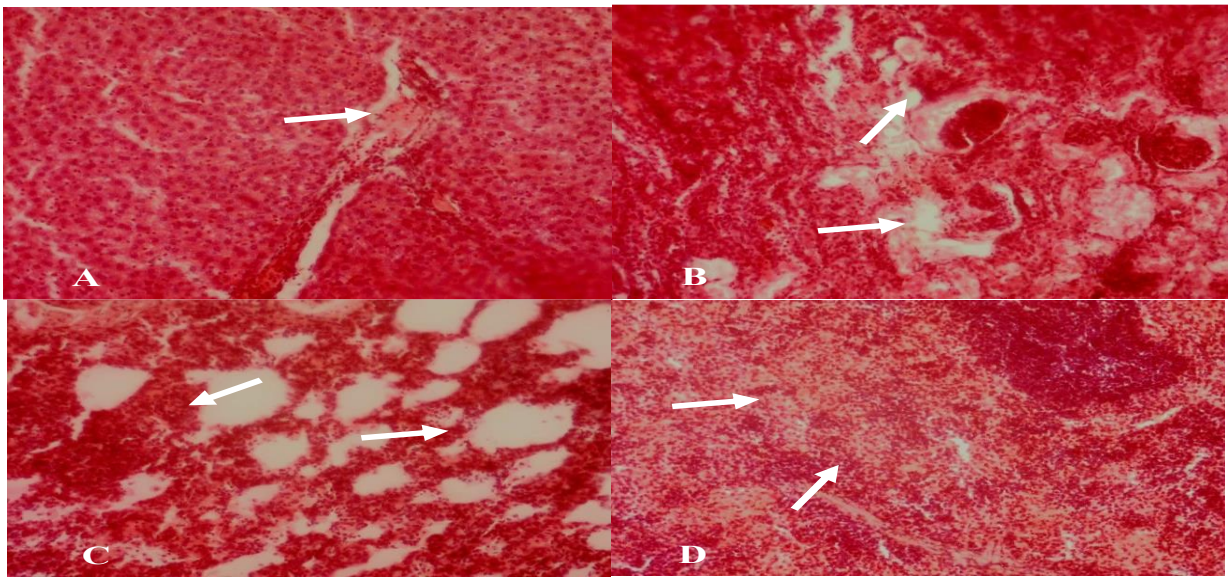

Plate 2: Photomicrographs of liver (A), kidney (B), lung (C), and spleen (D) from rats in negative control cadmium exposure. Note congestion (arrow) in A, necrotic tubules (arrows) in B, thickened interalveolar walls (arrows) in C; depletion of lymphoid cells in D; H & E × 100

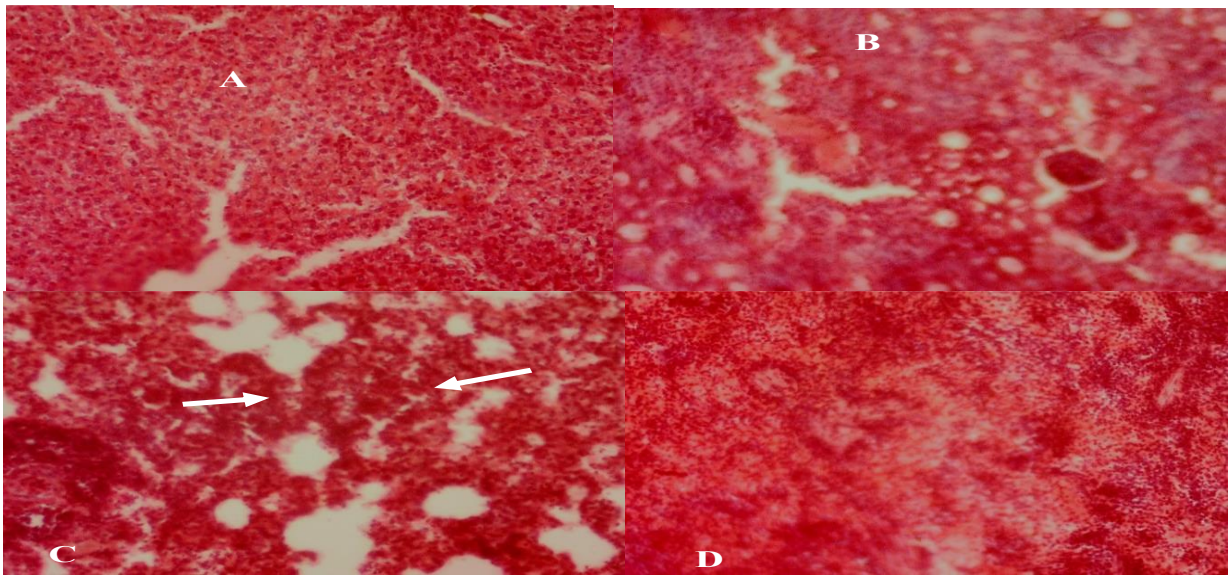

Plate 3: Photomicrographs of liver (A), kidney (B), lung (C), and spleen (D) from rats exposed to cadmium and treated with *Aloe barbadensis.* Note normal tissue architectures in A and D, relatively normal tissue in B, and thickened interalveolar walls (arrows) in C; H & E × 100

Conclusion

This research shows that *Aloe barbadensis* extract possesses some protective and metal-absorbing properties against cadmium toxicity in Wistar rats. The existence of functional groups such as carbonyl (C=O) and hydroxyl (O–H), as identified by FT-IR analysis, likely contributes to its ability to interact with and reduce cadmium bioavailability. Administration of the extract resulted in reduction in cadmium concentrations across examined tissues (0.01–0.03 mg/L) compared to the negative control groups (0.07–0.09 mg/L), indicating enhanced detoxification and reduced tissue accumulation. The findings suggest that *Aloe barbadensis* extract has promising potential as an all-natural medicinal substance to reduce cadmium-induced toxicity.

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Cite this article

Jangber Z.N., Ekwumemgbo P.A., Oyewale A.O., Garba Z.N. & Oladele S.B. (2026). Evaluation of Bioaccumulation of Cadmium in Wistar Rats and Absorption with *Aloe Barbadensis* Extract. FUAM Journal of Pure and Applied Science, **6**(**2**):83-90