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## THE PROTECTIVE AND CURRATIVE EFFECT OF *FICUS VOGELII* AGAINST TOXICITY INDUCED BY LEAD-ACETATE ON SPLEEN IN ADULT WISTAR RATS

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**ABSTRACT:** *Objective: This study aimed at investigating the protective and curative effect of Ficus vogelii against toxicity induced by lead acetate on spleen in adult Wistar rats. Methodology: Thirty (30) Wistar rats were used for this research and randomly assigned into 6 groups of five rats. Group A (Control group) received normal saline and water ad libitum. B was given lead acetate in a dose of 2mg/kg body weight for 28 days. The curative groups C (low dose) and D (high dose) received lead acetate and aqueous extract. The protective groups E (low dose) and F (high dose) received aqueous extract and lead acetate. Group G were given only aqueous extract. The experiment lasted for 28 days after 7 days of acclimatization. Results: Figure 1C showed lymphoid follicle with little necrosis demonstrating the effects of the extract as curative herb. Figure 1D shows expansion of red pulp and macrophages with relative improvement when compared to positive control. Figure 3E shows lymphoid follicle with necrosis and affected trabeculae (T). Figure 3F showed diffused red pulp and necrotized macrophages (arrows) which are not as much as that found in the positive control group and may be attributed to the protective role of the extract. The extract only (Figure 4) group shows normal histoarchitecture with red pulp, white pulp, microphages, trabeculae and lymphoid follicle all intact without any injury. Conclusion: This experiment demonstrates that lead causes splenic alterations like diffusion of white pulp into red pulp and appearance of large macrophages due to inflammations and production of debris of dead cells. These alterations can either be ameliorated or protect against by Ficus vogelii extract.*

**KEYWORDS:** Curative, Ficus Vogelii, Lead-Acetate, Protective, Toxicity, Spleen.

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### INTRODUCTION

Lead is an important metal with a bluish green colour. It is naturally available in small quantity as part of the earth's crust [1]. Lead is present in almost all parts of our environment such as water, air, food, soil and even fruits. Lead has found in the home for a very long time. Currently, it is used in the manufacturing of light-tech products for protections of nuclear reactors, thin sheets of electronic components, batteries, paints, ceramics, cables and ammunitions [2]. These products in our homes do not only cause health hazards to the people using them alone but due to their emissions, they pollute the environment causing health hazard to other inhabitants.

Lead gains entrance through various routes into the body via nasal, oral or dermal routes and cause intoxication [3,4]. It is very deleterious to almost all organs in the body and has

significant debilitating effects on the nervous, renal, hepatic, circulatory and hematopoietic system [5,6] while the two commonest routes of entry of lead into the body are inhalation and ingestion [7]. Ingestion is seeming to be more common source and inhalation is the most significant because pulmonary absorption is more efficient. They are various organs in the body that also show critical damage [8,9]. The ingested and absorbed lead are stored primarily in soft tissues and bone but the highest concentration of lead occurs in the bones, teeth, liver, lung, kidney, brain and spleen [10].

The spleen is the largest lymphoid organ with a very rich blood supply. The spleen is surrounded by a connective tissue capsule and divides its interior into incomplete compartments called the splenic pulp [11]. Being an important organ in the body in various ways, the spleen is concerned with phagocytosis and immune responses. In the fetus it is also an important site of haemopoiesis. Postnatally, it may become hematopoietic in some diseased conditions. Although very important in the defense of the body, it may not absolutely be essential because if the spleen is removed, many of its functions can be assumed by the liver and other lymphoid tissues [12,13]. With lead being so invasive, anything damage caused by the lead to the spleen is bound to affect all other organs that are supposed to take up the functions [14,15].

A lot of works has looked at the deleterious/toxic effects of lead on the histoarchitecture of spleen [16,17] and effects of other herbs and other substances on reducing the toxicity [18,19] but no researcher has ever considered the effect of *Ficus vogelii*. It is in view of the above that we designed this research work to evaluate the protective and curative effects of *F.vogelii* on the toxicity induced by lead acetate in the spleen of adult albino Wistar rats.

## **MATERIALS AND METHODS**

### **Experimental Protocol**

The thirty (30) healthy adult Wistar rats used for this research weighed between 180-200gm. They were acclimatized for 7 days and randomly assigned into 6 groups of five rats each. Group A (Control group) received normal saline and water *ad libitum*. Group B (experimental group) were given lead acetate in a dose of 2mg/kg body weight for 28 days. Group C (curative low dose) were given lead acetate and 100mg/kg of aqueous extract for 28 days. Group D (curative high dose) were given lead acetate and 300mg/kg of aqueous extract for 28 days. Group E (protective low dose) were given 100mg/kg of aqueous extract and lead acetate for 28 days. Group F (protective high dose) were given 300mg/k of aqueous extract and lead acetate for 28 days while Group G received 300mg/k of aqueous extract only for the same period. All the animals involved in this research received oral administration using orogastric tube [20].

### **Ethical Clearance**

During this experiment, we respected and strictly observed the following councils' directive on the use of experimental animals. They are; Directive 2010/63/Eu, 2010 of the European parliament and the European Council as passed on 22 September, 2010 on the use and protection of experimental animals [21] and the Organization of Economic Co-operation Development, Paris, guideline for testing of chemical usage in Experimental animals, OECD,

[22]. These animals were fed with standard rat feed and allowed water *ad libitum* [19,23]. We also sort for and obtained the ethical clearance from the Faculty of Basic Medical Sciences University of Nigeria Enugu Campus.

### **Leaves Collection and Extraction**

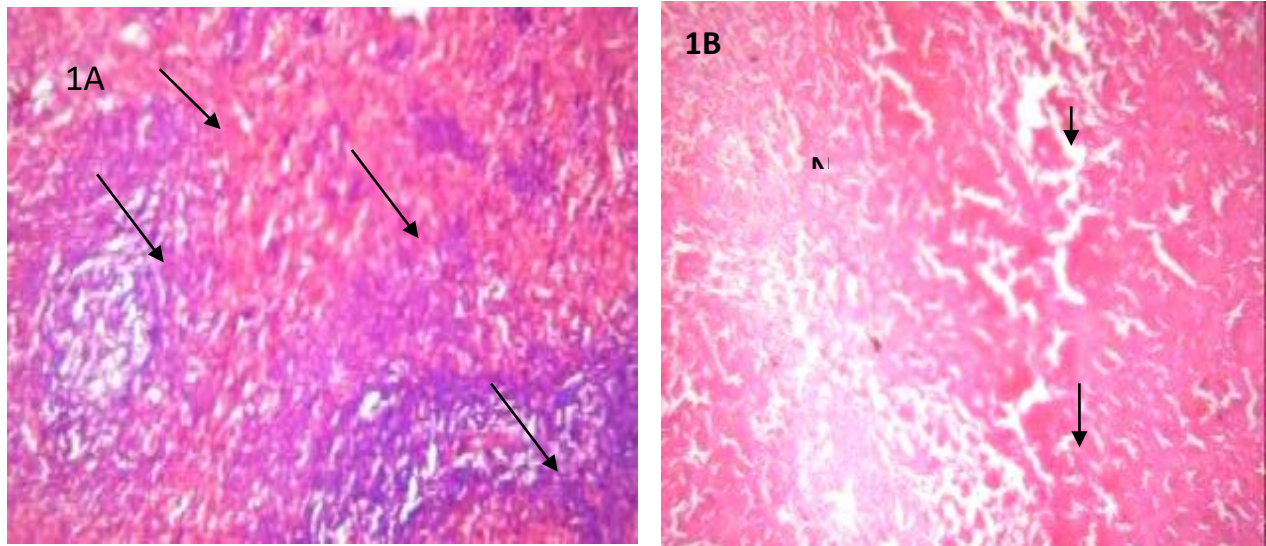
The fresh leaves of *F. vogelii* were collected from Enyibichiri Ndufu-Alike Ikwo in Ikwo Local Government Area of Ebonyi State. These leaves were washed and dried in a ventilated room. Thereafter, it was crushed into powder using electronic blender and passed through mesh sieve to get the fine powders. Five hundred grams (500g) of the powder was weighed using an electronic weighing balance and soaked in 1200mL of water (powder/solvent). The mixture was agitated using an electric blender to enhance proper mixing of the solvent with the powder and then poured into air-tight plastic containers. The container with the mixture was kept in a refrigerator for 48hours [24]. The mixtures were filtered first with cheese cloth, and then with Whiteman No 1 filter paper (24cm). The filtrates were separated and concentrated in vacuum using Rotary Evaporator to 10% of their original volumes at 37<sup>o</sup>C - 40<sup>o</sup>C. These were concentrated using a water bath until a sticky paste was gotten. The extracts were stored in a refrigerator until it is required for use. All preparations were performed at the Department of Anatomy Faculty of Basic Medical Sciences, College of Medicine University of Nigeria Enugu Campus (UNEC).

### **Histological Study**

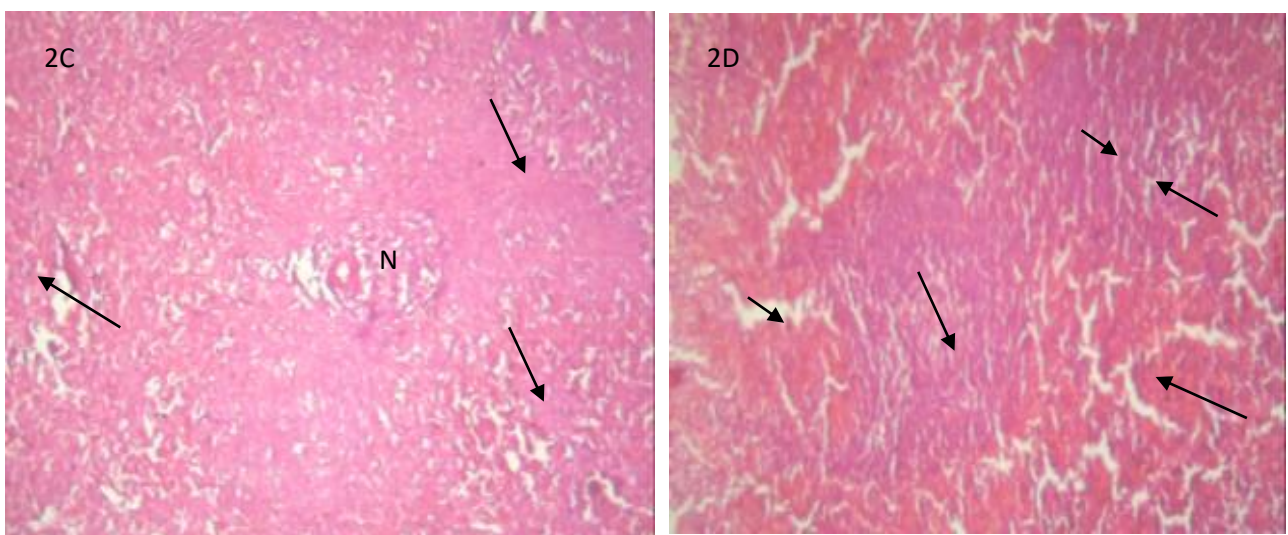
At the end of the experiment, the rats were starved for 24 hours and anaesthetized and then decapitated [25]. The animals were dissected and the liver removed and quickly fixed in 10 % formalin for routine histological procedures. The tissues were processed and embedded in paraffin wax. Thin sections of 4-5 $\mu$ m were obtained and stained using haematoxylin and eosin (H&E) and were examined under light microscope to determine the histological changes in the liver histology.

## RESULTS

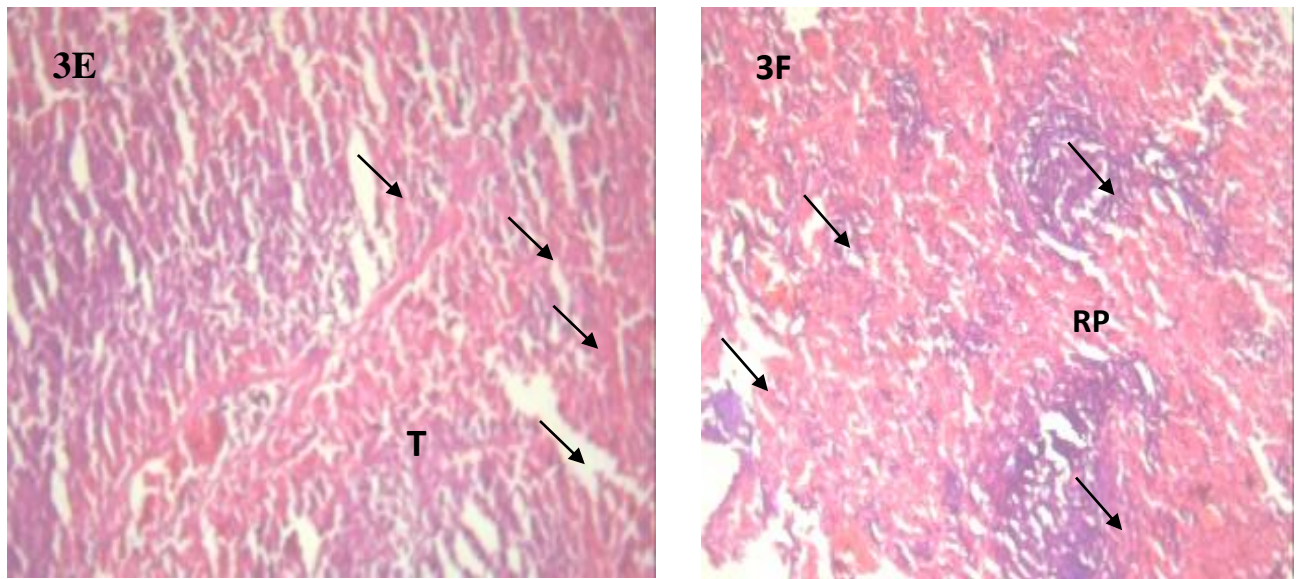
The results of the experiment are presented below as a photomicrographic plate. These sections were taken after a very routine histological process had been undertaken.



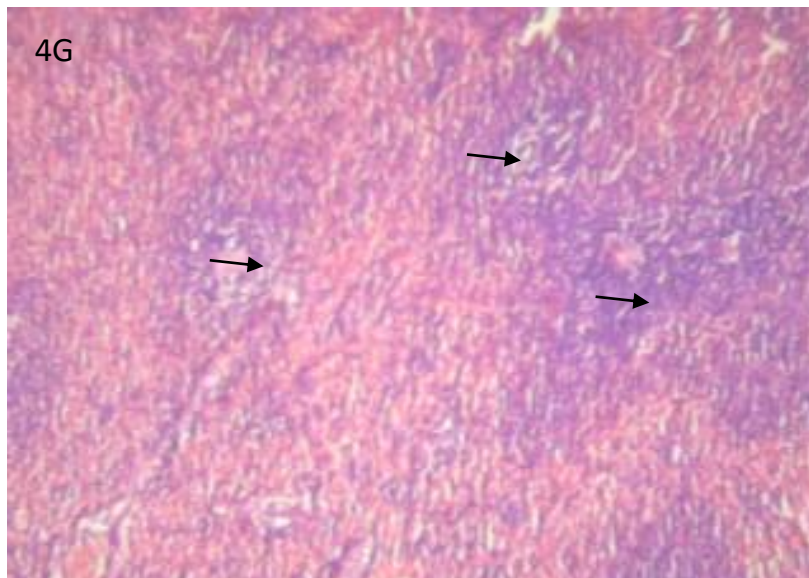
**Figure.1: Photomicrograph of rat; (A) spleen that received only normal saline showing normal white pulp, red pulp and normal spleen histology, H & E, Mag; 200. (B) Photomicrograph of rat that received only lead acetate showing N-necrotized area, diffused white pulp, distorted red pulp and damaged histology, H & E, Mag; 200.**



**Figure 2: Photomicrograph of rat; (C) spleen treated with lead acetate before *F.vogelii* 100mg/kg showed lymphoid follicle without necrosis (H&E-Mag, 200). (D) Spleen treated with lead acetate before *F.vogelii* 300mg/kg showed expansion of red pulp and macrophages (arrows) (H&E-Mag, 200).**



**Figure 3: Photomicrograph of rat; (E) spleen treated with *F.vogelii* 100mg/kg before showed lymphoid follicle with necrosis and affected trabeculae (T) (H&E-Mag, 200). (F) Spleen treated with *F.vogelii* 300mg/kg before lead acetate showed diffused of red pulp and necrotized macrophages (arrows) (H&E-Mag, 200).**



**Figure 4: Photomicrograph of rat spleen that received only 300mg/kg of the aqueous extract showing normal histological architecture with red pulp, white pulp, macrophages and lymphoid follicle.**

## DISCUSSION

Human health has been in danger recently due to the emissions from our various home appliances as a result of science and technology and from our natural environment. These emissions we may not be in control of but we can either manage the way we interact with them or use our natural products such as the herbs that have been proven to be antitoxicity to counteract their effect. After the present research, the histological investigations of the spleens in the normal control group were not affected as it did not receive anything other than water and feed. All the treated rats showed distortions in their splenic architectonics such as the red and white pulp besides fibrous capsule as a covering for the spleen [26]. According to Aldahmash and El-Nager, [27] the red pulp is the area of spleen in between white pulp and consists of open sinuses and cellular cords. Splenic sinuses are open vascular spaces lined by a discontinuous layer of endothelial cells and supported by a fenestrated basal lamina and reticular fibers. The surrounding cellular splenic cords provide a tissue frame work maintaining the network of sinuses.

The positive control that was treated with lead acetate only showed that the spleen of the rats presented several distortions that represent the deleterious effects of lead on the spleen architecture [28]. The white pulp was caused to diffuse into the red pulp which lead to the two structures being ill defined. There also various areas that showed necrosis as a result of the induced lead acetate toxicity [27]. The cells mostly responsible for slowing the propagation of invading pathogen in the spleen are macrophages and polymorphonuclear (phagocytes cells), these cells are in charge while an antigen-specific adaptive immune response (antibody- or cell-mediated) is being established. Various authors such as Kowolenko *et al.*, [29]; Mael *et al.*, [30] reported that lead can inhibit macrophage function possibly by overloading macrophages with cellular debris and inhibiting macrophage production of nitric oxide [31].

The photomicrograph of spleen in figure 1C spleen treated with lead acetate before *F.vogelii* 100mg/kg showed lymphoid follicle without much necrosis which may be demonstrating the effects of the extract as curative herb. Figure 1D is showing the spleen of rats treated with lead acetate before *F.vogelii* 300mg/kg with expansion of red pulp and macrophages (arrows) which showed relative improvement when compared with the positive control. Figure 3E represents the spleen of rats treated with *F.vogelii* 100mg/kg before lead acetate showing lymphoid follicle with necrosis and affected trabeculae (T). in figure 3F, the Spleen of the rats treated with *F.vogelii* 300mg/kg before lead acetate showed diffused red pulp and necrotized macrophages (arrows) which are not as much as that found in the positive control group and may be attributed to the protective role of the extract. The results of the present study agreed with those of Aly *et al.* [32] who showed that the effects of lead toxicity in spleen can be countered. Photomicrograph of rat spleen that received only 300mg/kg of the aqueous extract (Figure 4) shows normal histoarchitecture with red pulp, white pulp, microphages, trabeculae and lymphoid follicle all intact without any injury.

Our present experiment demonstrates that oral administration of lead results in several alterations in the spleen represented by diffusion of white pulp into the red pulp and appearance of large macrophages due to the inflammations caused by toxicity and production of debris of dead cells. This agrees with the report of Aldahmash And El-Nager, [27].

## CONCLUSION

The herb did a great work in the restoration of the integrity of the spleen histology that is to say it act well as a curative medicine. The extract was also good as a protective herb. Comparing the both the herb could better be used as a protective medicine because it tends to protect more than when used as curative medicine.

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