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Effect of Ethanolic Leaf Extract of *Telfairia occidentalis* on the Histology of Kidneys of Lead-Induced Wistar Rats

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Abstract:

Objective: This research study was carried out to investigate the effect of ethanolic leaf extract of *Telfairia occendentalis* on the histology of kidneys of lead-induced wistar rats.

Methodology: Twenty-five (25) male wistar rats weighing 150-180g were procured and acclimatized for two weeks, after which, they were divided into five (5) groups of five (5) rats each, and were housed in cages. The groups were designated as groups A - E. Group A served as the control group and was not induced with lead (Pb), while Groups B – E were induced. Groups A received distilled water only, Groups B - E received vitamin C, vitamin C + 100mg/kg of ethanolic leaf extract *Telfairia occendentalis,* vitamin C + 400mg/kg ethanolic leaf extract of *Telfairia occendentalis* and vitamin C + 800mg/kg ethanolic leaf extract of *Telfairia occendentalis* respectively for 14 days through oral route with the aid of oral gastric tube. On the 15th day, the animals were weighed and sacrificed via chloroform inhalation, and kidneys were harvested from the rats for histological study.

Results: Histopathological findings showed normal renal architecture with glomeruli (G), renal tubules (RT), tubular cells (TC), and normal cuboidal epithelial cells(CEC) within the medullar for animals in group A; moderate degeneration with moderate fatty changes (FC), moderate intra renal hemorrhage (IRH), and moderate renal inflammation (IRI) for animals in group B; mild regeneration with moderate fatty changes (FC), tubular atrophy (TA), and moderate renal inflammation (IRI) for animals in group C; moderate regeneration with mild fatty changes (FC), and mild renal inflammation (IRI) for animals in group D; and moderate regeneration with mild fatty changes (FC) otherwise normal with well outlined tubular cells (TC) for animals in group E.

Conclusion: Ethanolic leaf extract of *Telfaria occendentalis* have ameliorating effect on the histology of kidneys of lead-induced wistar rats, and the ameliorating effect improves with increase in the dosages of the leaf extract.

Keywords: *Telfaria occendentalis*, Kidneys, Lead.

I. Introduction

Lead exposure is estimated to accounts for 21.7 million years lost to disability and death worldwide due to long-term effects on health, with 30% of the global burden of idiopathic intellectual disability, 4.6% of the global burden of cardiovascular disease and 3% of the global burden of chronic kidney diseases [1]. its exposure is a global issue since its mining and smelting, and battery manufacturing, disposal, and recycling are common in many countries [2]. It has no confirmed biological role, and no confirmed safe level of lead exposure [3]. It is a highly poisonous metal (whether inhaled or swallowed), affecting almost every organ and system in the human body [4]. At airborne levels of 100 mg/m^3, it is immediately dangerous to life and health [5]. Most ingested lead is absorbed into the bloodstream [6]. The primary cause of its toxicity is its predilection for interfering with the proper functioning of enzymes for which it does by binding to the sulfhydryl groups found on many enzymes [7], or mimicking and displacing other metals which act as cofactors in many enzymatic reactions [8]. Essential metals that lead interacts with include calcium, iron, and zinc [9]. High levels of calcium and iron tend to provide some protection from lead poisoning; and low levels cause increased susceptibility [10]. Research has shown that lead can cause severe damage to the brain and kidneys, and, ultimately, death [2]. According to WHO [1] once lead enters the body, it is distributed to organs such as the brain, kidneys, liver. and bones. The body stores lead in the teeth and bones, where it accumulates over time, and lead stored in bone may be released into the blood during pregnancy, thus exposing the fetus. Undernourished children are more susceptible to lead because their bodies absorb more lead if other nutrients, such as calcium or iron, are lacking, and children at highest risk are the very young (including the developing fetus) and the economically disadvantaged [2].

ISSN No. 231-2705 | DOI: 10.51244/IJRSI | Volume X Issue VI June 2023

Symptoms of lead poisoning include nephropathy, colic-like abdominal pains, and possibly weakness in the fingers, wrists, or ankles [2]. Small blood pressure increases, particularly in middle-aged and older people, may be apparent and can cause anemia [2]. Several studies, mostly cross-sectional, found an association between increased lead exposure and decreased heart rate variability [11]. In pregnant women, high levels of exposure to lead may cause miscarriage, and chronic, high-level exposure has been shown to reduce fertility in males [12]. Mycyk *et al*, [13] stated that in a child's developing brain, lead interferes with synapse formation in the cerebral cortex, neurochemical development (including that of neurotransmitters), and the organization of ion channels. Early childhood exposure has been linked with an increased risk of sleep disturbances and excessive daytime drowsiness in later childhood [14]. The rise and fall in exposure to airborne lead from the combustion of tetraethyl lead in gasoline during the 20th century has been linked with historical increases and decreases in crime levels. Medicinal plants which are composed of phytochemicals and phytonutrients, possess therapeutic properties, or exert beneficial pharmacological effect on the human body and animal body; and include such plants such as *Telfaria occendentalis* that could help to improve the health of diseased living organisms.

Telfairia occidentalis is a tropical vine grown in West Africa as a leaf vegetable, and for its edible seeds. Its common names include fluted gourd, fluted pumpkin, ugu (in the Igbo language), okwukwo-wiri (in Ikwerre language) and ikong-ubong (in the Efik and Ibibio languages) [15]. It is a member of the family Cucurbitaceae, and is indigenous to southern Nigeria [16]. It grows in many nations of West Africa, and is mainly cultivated in southeastern Nigeria. It is used primarily in soups and herbal medicines [17]. *T. occidentalis* is traditionally used by an estimated 30 to 35 million people in Nigeria, including the Efik, Ibibio, Ikwerre, and Urhobo ethnic groups [16], However, it is predominantly used by the Igbo ethnic group, who continue to cultivate the gourd for food sources and traditional medicines [18].

The nutritional components of *T. occidentalis* (Ugu) leaf include calcium, potassium, iron and folic acid, vitamin E and B6, magnesium, phosphorus, thiamine, niacin, copper, manganese, protein, vitamin C, riboflavin, and dietary fiber [19]. Considered an "oil seed" the fluted gourd is high in oil (30%) [16]. Shoots of *T. occidentalis* contain high levels of potassium and iron, while seeds are composed of 27% crude proteins and 53% fats [20]. The leaves contain a high quantity of antioxidants and hepatoprotective and antimicrobial properties [17]. The young shoots and leaves of the female plant are the main ingredients of a Nigerian soup, *ofe egwusi*. The large (up to 5 cm), dark-red seed is rich in fat and protein and can be eaten whole, ground into powder for a kind of soup, or made into a fermented porridge [15].The edible seeds can be boiled and eaten whole, or fermented and added to *ogili* [21].

T. occidentalis has been traditionally used by indigenous tribes as a blood tonic, likely due to its high protein content [16]. Flour produced from the seeds can be used for high-protein breads [22]. Furthermore, the shoots and leaves can be consumed as vegetables [10]. When *T. occidentalis* is prepared for herbal medicine, it is used to treat sudden attack of convulsion, malaria, and anaemia; it also plays a vital and protective role in cardiovascular diseases [23]. Study has shown that its leaf enhances blood production, serve as excellent source of dietary fiber, function as anti-diabetic agent, protects body's tissues, and strengthens bones teeth [19]. Other benefits include treatment of convulsion, antioxidant-rich, hormone harmonization, weight loss effective, helps fertility, lactating characteristics, anti-inflammatory effects, antimicrobial properties, treatment of anaemia, lowers the risk of kidney disease, helps to prevent cancer, antioxidant abilities, stress relief, prevents Alzheimer's disease and dementia, treats infertility and immune system booster [19]. According to Okoye *et al*, [24], leaf extract of *T. occidentalis* might lead to membrane stabilizing effects on hepatocytes, depressed hepatocyte synthetic activity and impaired renal function, and has effectively maintain electrolyte balance, modulates pancytopenia and oxidative renal damage in rats suggesting its protective potentials on anaemia and renal disorders [25]. It is also a better blood boosting vegetable than *A. conyzoides* [26].

Thus, this study will help to create more awareness on the ameliorating effect of the leaf extract of *Telfairia occidentalis* on lead related poisoning thereby encourage regular consumption of the leaf to reduce effects of lead poisoning which affects almost all the organs of the body.

II. Materials and Methods

2.1 Animal procurement, care, and treatment

Fwenty-five (25) male wistar rats weighing between 150g to 180g were procured and housed at the Animal house of Anatomy Department, Abia State University; Uturu with wire gauze cages in a well-ventilated area, were maintained under standard laboratory conditions of temperature ($22+2^0$C), relative humidity (55-65%) and 12 hours light/dark cycle. They were fed with standard commercial pellet diet and water *ad libitum* and were also acclimatized for two weeks before the experiment. Their health statuses were closely monitored before and during the experiment. All procedures were carried out in strict accordance with the Institutional guidelines on the care and use of experimental animals.

ISSN No. 231-2705 | DOI: 10.51244/IJRSI | Volume X Issue VI June 2023

2.2 Collection, identification, and preparation of plant material

Fresh leaves of *Telfairia occidentalis* were purchased from a local market in Okigwe L.G.A., Imo State, and were authenticated at Herbarium unit, Botany Department, Abia State University, Uturu, Abia State. The leaves were air dried and crushed using laboratory blender. Extractions were done using ethanol. The crude ethanol extracts were kept in an air-tight container and stored in a refrigerator at 4^0C until time of use. At the time of use, the ethanol extracts were filtered into a stainless basin with a white cloth and placed in a water bath to dry up the ethanol. 250mg of these extracts /kg body weights were dissolved in 10mls of distilled water and were administered to the animals.

2.3 Induction of lead and Vitamin C administration

According to research study, the oral LD50 of lead acetate has been calculated to be 600 mg/kg body weight for Wistar rats [27], thus, 5% (30 mg/kg) of the lead acetate were used to induced the rats daily for fourteen (14) days. Also, 40mg/kg body weight of vitamin C was administered to the lead-induced animals for the same number of days.

2.4 Experimental protocol

The animals were grouped into five (5) groups of five (5) rats each. Different doses of the leaf extracts were administered via oral route with the aid of oral gastric tube as shown below:

Group A The control group + distilled water.

Group B Lead + Vitamin C

Group C Lead + Vitamin C + 100mg/kg of *T. occidentalis* leaf extract.

Group D Lead + Vitamin C + 400mg/kg of *T, occidentalis* leaf extract.

Group E Lead + Vitamin C + 800mg/kg of *T. occidentalis* leaf extract.

2.5. Sample collection and analysis

The extracts were administered for fourteen (14) days. On the 15th day, the animals were sacrificed by anaestethizing under chloroform vapour and dissected. Kidneys harvested from the wistar rats, weighed, and were fixed in Bouin's fluid for 72 hours, after which they were transferred to 10% buffered formalin. This was followed by histological and histochemical methods of tissue processing.

III. Results

3.1. Histopathological findings

Micrograph 1 is the result of the microscopic examination of the kidneys of the animals in group A (1R1R2) (x400) (H/E) showing normal renal architecture with glomeruli (G), bowman space (BS), renal tubules (RT), tubular cells (TC), and normal cuboidal epithelia cells (CEC) within the medullar.

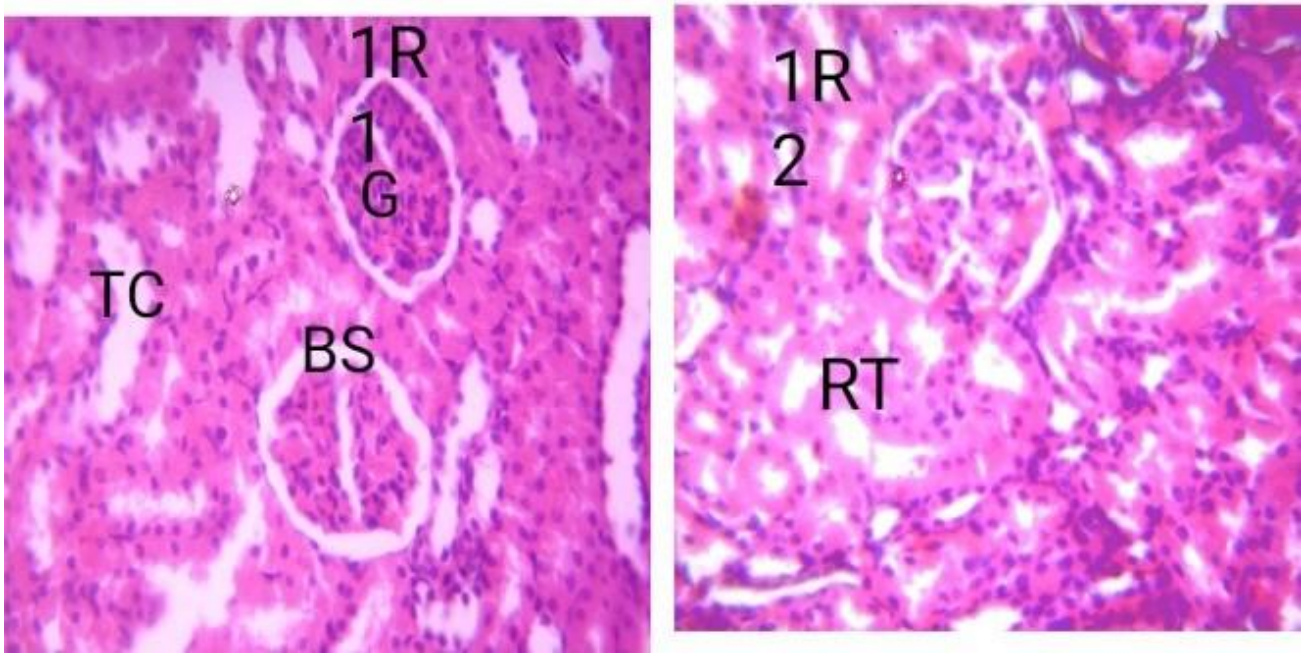
Micrograph 2 is the result of the histology of the kidneys of the animals in group B (2R1R2) induced with lead (x400) (H/E) treated with Vitamin C showing moderate degeneration with moderate fatty changes (FC), moderate intra renal hemorrhage (IRH) and moderate renal inflammation (IRI).

Micrograph 3 is the result of the histology of the kidneys of the animals in group C (3R1R2) induced with lead (x400) (H/E) treated with Vitamin C + 100mg/kg of *T. occidentalis* leaf extract showing mild regeneration with moderate fatty changes (FC), tubular atrophy (TA), and moderate renal inflammation (IRI).

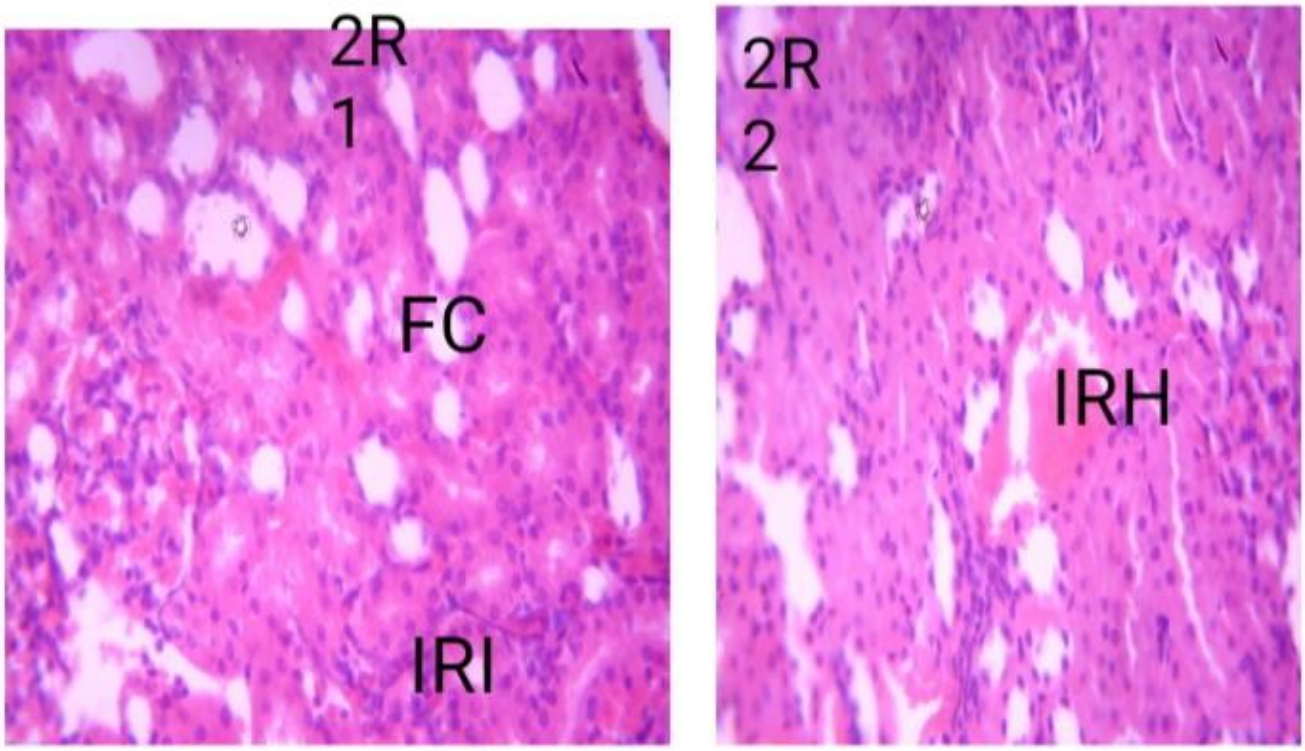
Micrograph 4 is the result of the histology of the kidneys of the animals in group D (4R1R2) induced with lead (x400) (H/E) treated with Vitamin C + 400mg/kg of *T. occidentalis* leaf extract showing mild regeneration with mild fatty changes (FC), and mild renal inflammation (IRI).

Micrograph 5 is the result of the histology of the kidneys of the animals in group E (5R1R2) induced with lead (x400) (H/E) treated with Vitamin C + 800mg/kg of *T. occidentalis* leaf extract showing moderate regeneration with mild fatty changes (FC), otherwise normal with well outlined tubular cells (TC).

ISSN No. 231-2705 | DOI: 10.51244/IJRSI | Volume X Issue VI June 2023

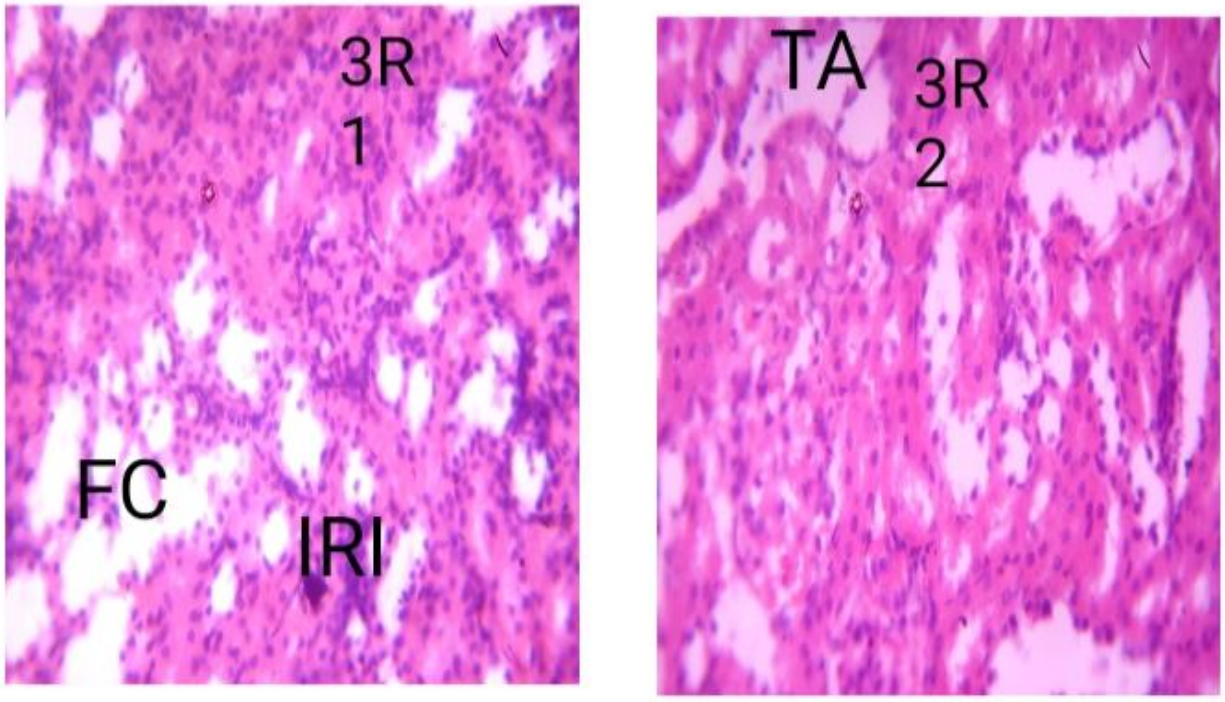


Micrograph 1 showing normal renal architecture with glomeruli (G), bowman space (BS), renal tubules (RT), tubular cells (TC), and normal cuboidal epithelia cells (CEC) within the medullar.

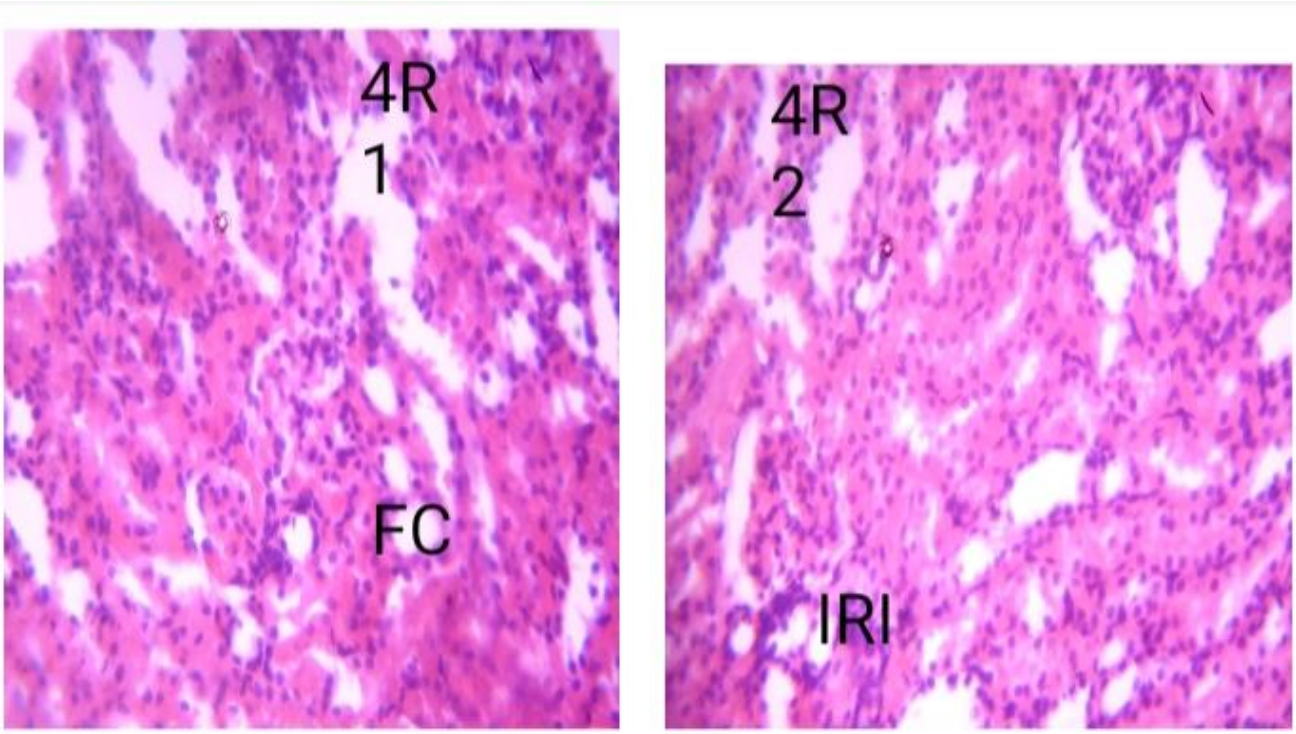


Micrograph 2 is showing moderate degeneration with moderate fatty changes (FC), moderate intra renal hemorrhage (IRH) and moderate renal inflammation (IRI).

ISSN No. 231-2705 | DOI: 10.51244/IJRSI | Volume X Issue VI June 2023

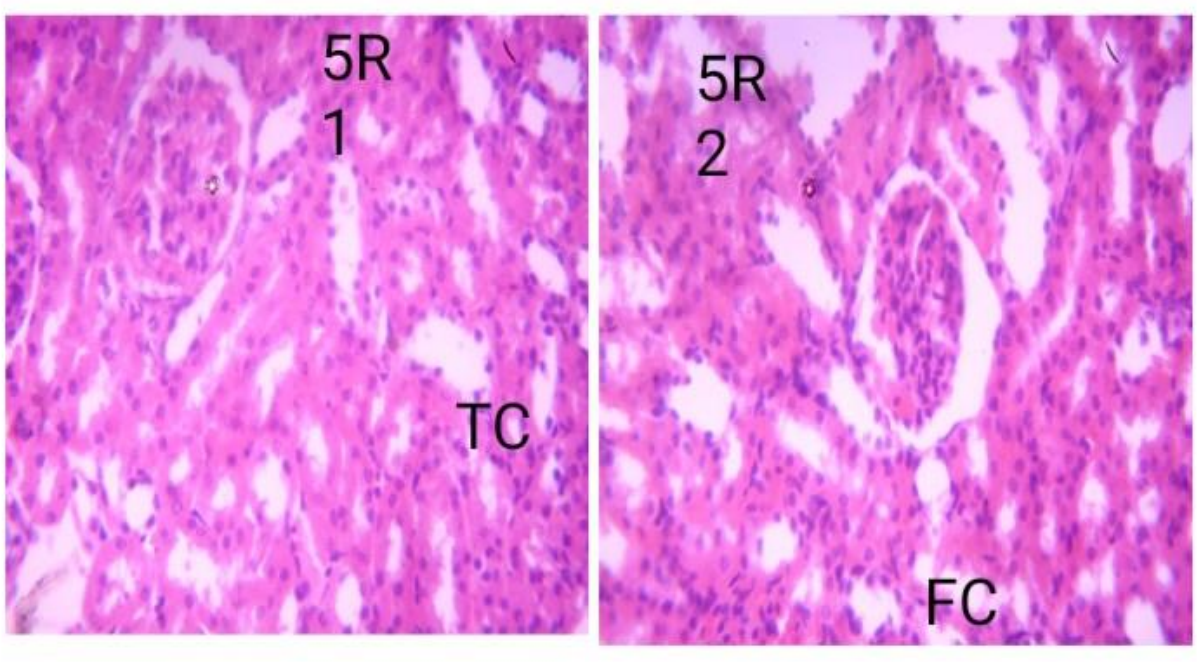


Micrograph 3 is showing mild regeneration with moderate fatty changes (FC), tubular atrophy (TA), and moderate renal inflammation (IRI).



Micrograph 4 is showing mild regeneration with mild fatty changes (FC), and mild renal inflammation (IRI).

ISSN No. 231-2705 | DOI: 10.51244/IJRSI | Volume X Issue VI June 2023



Micrograph 5 is showing moderate regeneration with mild fatty changes (FC), otherwise normal with well outlined tubular cells (TC).

IV. Discussion

According to Tayaba, [28], lead is highly persistent in the environment, and its continuous use raise the level of lead in every country, causing serious threats such as carcinogenicity, renal failure, high blood pressure, brain damage, hematological effects, reproductive system damage both in men and women, heart diseases, bone screening, liver damage etc. Tayaba [28] further stated that Lead has no beneficial effects on human health, thus, leads to several complications like anemia, carcinogenicity, damage of reproductive system of men and women, kidney damage, heart disease, brain damage, raised of blood pressure, liver damage, effects on children learning ability and behavior etc. Due to increased human activities the amount of lead has increased in the environment, thus failure to control the level of lead will lead to severe complications in future [28].

The histopathological finding of this present study of group A (1R1R2) (x400) (H/E) showed normal renal architecture with glomeruli (G), bowman space (BS), renal tubules (RT), tubular cells (TC), and normal cuboidal epithelia cells (CEC) within the medullar due to non-exposure to lead acetate. While, the histopathological result of the histology of the kidneys of the animals in group B (2R1R2) induced with lead (x400) (H/E) treated with Vitamin C showing moderate degeneration with moderate fatty changes (FC), moderate intra renal hemorrhage (IRH) and moderate renal inflammation (IRI). This could be due to lead toxicity interfering with the proper functioning of enzymes by binding to the sulfhydryl groups found on many enzymes [7], or mimicking and displacing other metals which act as cofactors in many enzymatic reactions [8]. According to Nigra *et al*, [29] exposure to high levels of lead can result in adverse health outcomes, including damage to the nervous system, liver and kidneys, anaemia, hypertension, cardiovascular disease, immune deficiency, infertility, developmental problems including cognitive deficits, learning disability and memory loss. Moreso, lead can cause severe damage to the brain and kidneys, and, ultimately, death [2], and once lead enters the body, it is distributed to organs such as the brain, kidneys, liver, and bones [1].

Micrographs 3, 4 and 5 of the histology of the kidneys of the animals in groups C, D, and E that were induced with lead and were given Vitamin C + 100mg/kg, Vitamin C + 400mg/kg, and Vitamin C + 800mg/kg of *T. occidentalis* leaf extracts showed mild regeneration with moderate fatty changes (FC), tubular atrophy (TA), and moderate renal inflammation (IRI); mild regeneration with mild fatty changes (FC), and mild renal inflammation (IRI); and moderate regeneration with mild fatty changes (FC), otherwise normal with well outlined tubular cells (TC) respectively may be due the ameliorating effect of *T. occidentalis* leaf extracts. Research study has shown that the benefits of *T. occidentalis* leaf include treatment of convulsion, antioxidant-rich, hormone harmonization, weight loss effective, helps fertility, lactating characteristics, anti-inflammatory effects, antimicrobial properties, treatment of anaemia, lowers the risk of kidney disease, helps to prevent cancer, antioxidant abilities, stress relief, prevents Alzheimer's disease and dementia, treats infertility and immune system booster [19]. Also, leaf extract of *T. occidentalis* might lead to membrane stabilizing effects on hepatocytes, depressed hepatocyte synthetic activity and impaired renal function,

ISSN No. 231-2705 | DOI: 10.51244/IJRSI | Volume X Issue VI June 2023

and has effectively maintain electrolyte balance, modulates pancytopenia and oxidative renal damage in rats suggesting its protective potentials on anaemia and renal disorders [25].

Since the essential metals that lead interacts with include calcium, iron, and zinc [9], and research has shown that high levels of calcium and iron tend to provide some protection from lead poisoning; and low levels cause increased susceptibility [10], it therefore means that the ameliorating effect of *T. occidentalis* (Ugu) leaf may have come from its nutritional components include calcium, potassium, iron and folic acid, vitamin E and B6, magnesium, phosphorus, thiamine, niacin, copper, manganese, protein, vitamin C, riboflavin, and dietary fiber (30-amazing-benefits-of-ugu). However, its effect is dose-dependent with better ameliorating effect at the histology of the kidneys of the animals in group E that received vitamin C + 800mg/kg of *T. occidentalis* leaf extracts.

V. Conclusion

Leaf extract of *Telfaria occendentalis* has ameliorating effect on the histology of kidneys of lead-induced wistar rats, and the ameliorating effect is dose-dependent, and improves better with increase in dosages of the leaf extract.

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Disclosure of conflict of interest

No conflict of interest.

Statement of ethical approval

Approved by Institutional ethical approval.

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