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Histological, Hematological and Biochemical Study of Some Siqua Constituents That Effect on Some Organs in Male Albino Mice.

A Thesis Submitted in Partial Fulfillment of the Requirements for the Master's Degree of Science in Biology

By

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بِسْمِ اللهِ الرَّحْمنِ الرَّحِيمِ

﴿ فَتَعَالَى اللَّهُ الْمَلِكُ الْحَقُّ وَلَا تَعْجَلْ بِالْقُرْآنِ مِنْ قَبْلِ

أَنْ يُقْضَى إِلَيْكَ وَحْيُهُ وَقُلْ رَبِّ زِدْنِي عِلْمًا ﴾

صدق الله العلي العظيم

سورة طه: الآية "١١٤"

Dedication

I dedicate all my efforts to

Whom I worship for His merciful

Allah

The leaders of my Life Prophet

Mohammed and His family

To whom did not spare anything to push me on the path of success, who taught me to ascend the ladder of life with wisdom and patience

Soul of my dear father

To the one who weaved my happiness with threads woven from her heart

My mother.

To whom braced me throughout my studies, my darling wife

Duaa

To the flowers of Jasmine and sweetheart my children

Fatima, Ruqaya, Abdullah

To those who have watered ambition in myself

my sisters & brother

To everyone helped and supported me to do this work……
I introduce my work with love & respects

Bashar Abdullah

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Abstract

The current study focused on the histological and biochemical change in some organs of male white mice after treated with lead and *Nerium oleander* as main structures of siqua. Studied animals were divided into four groups, three of them treated as groups (B, C and D) each group was treated with one important structure of siqua, while the group A that considered as the control group.

The main histological results included aggregation of inflammatory cells and sever degeneration in brain tissue. The histological section of brain after treated with *Nerium oleander* showed hypertrophy in nerve cells, blood hemorrhage, blood congestion and necrosis in brain tissue, so, showed Apoptosis in the different regions of brain. However, the brain tissue after treated with lead and *Nerium oleander* noted hemolysis spots, cellular proliferation of inflammatory cells. The histological change in small intestine after being treated with lead showed prominent destruction in mucosa layer and the epithelial cells loss their nuclei, in the lumen of intestine appeared after treated with *Nerium oleander* exfoliated in epithelial layer and muscularis was spirated into three thin, while the result of intestine after treated with lead and *Nerium oleander* showed completely the separated the submucosal layer from tunica muscularis and destruction villi.

The histological result of liver after treated with lead and *Nerium oleander* noted parenchyma have hemorrhages, prominent necrosis lesions and the liver tissue acute cellular destruction ,liver have wide circular space contained on RBC respectively. The liver after treated with compound lead and *Nerium oleander* appeared abnormal portal area and filled with blood.

The result of kidney after treated with lead showed destruction in the glomerular capillaries and blood hemorrhage, the tissue section of kidney after treated with *Nerium oleander* and lead and *Nerium oleander* showed tissue destruction in the cortical region, acute degeneration in the wall of P.C.T and D.C.T. and abnormal cellular proliferation in the cortex and renal corpuscles have prominent destruction in the glomerular capillaries respectively. The biochemical result of (AST, ALT, urea, creatinine and C.B.P) after three treated groups with (*Nerium oleander*, lead and lead+*Nerium oleander*) showed significant differences compared with control group.

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List of Abbreviations

Abbr.	Abbreviations
ATSDR	Agency for Toxic Substances and Disease Registry
EPA	Environmental Protection Agency's
USEPA	United States Environmental Protection Agency
WCSP	World Checklist of Selected Plant Families
A.P	Associate Press
ROS	Reactive oxygen species
WHO	World Health Organization
USDA	United States Department of Agriculture
ALT	Alanine Transaminase
AST	Aspartate Transaminase
ALP	Alkaline phosphatase
P.C.T	Proximal Convoluted Tubule
D.C.T	Distal Convoluted Tubule
D.B	Descending Branch
A.B	Ascending Branch
C.D	Collecting Duct
EDTA	Ethylene Diamine Tetraacetic Acid
WBCs	White Blood Cells
RBCs	Red Blood Cells
HGB	Hemoglobin
PLTs	Platelets
LYM	Lymphocytes
DALYs	Disability-adjusted life years
IHME	Institute for Health Metrics and Evaluation

Introduction

Siqua refers to a mix of a some material used a medicine for children which infected with diarrhea used by people with little knowledge who live in countryside where the Siqua is common as a drug, causing a lot of danger effects on people who got it.

This Siqua compassion have three sources which include :

1) Mineral sources : Lead _ Ink .
2) Planet sources : *Nerium oleander*.
3) Animal sources : Skin of Urchin_ Nails of either human or animal.

Lead was conceded as a bluish-gray, solid metal, heavy, low melting, it's density at 20˚C is 11.34 g/cm³, chemical formula is Pb (comes from the Latin word "Plumbum"), boiling point 1,740C˚, melting point 327.4C˚, insoluble in water, and molecular weight 207.20 (AIC, 2007).

Lead (Pb) is one of the major toxic heavy metals; it is arranged under 275 hazardous matters according to Environmental Protection Agency's (EPA) Priority List of Hazardous Substances formulated by the Agency for Toxic Substances and Disease Registry (ATSDR) (Stevens *et al*., 2002). Pb is not biodegradable, it present in environment, becoming a source of pollution and risk to human life and other living organisms (Hariharan *et al*., 2016). Lead documented as chronic and acute intoxication for animals and humans , it is used in industrial activities, but exposure to lead by food or water and air pollution in many countries continues and become a universal problem, pollution caused by industrial emission and combustion of lead including gasoline (Mudipalli, 2007).

Exposure to lead causes neurotoxic effects including neurodevelopment, disturbed light dark preference, and neuro-behavioral alteration (Li *et al*., 2019). Poisoning exerts most severe consequences in the developing brain due to the immature blood-brain barrier and the absence of protein complexes able to sequester lead in mature tissue (Goyer, 1990).

Toxicity of lead was investigated on some organs like nephrotoxic, the effect exhibited with microscopic examination which showed severe changes on kidney scarping and renal tubules (Loumbourdis, 2003). Lead cause blood symptoms and decrease in blood levels, cardiova causes colic, and blood gastrointestinal

immunotoxicity, growth deficits and development, hypertension, anemia, in addtion lead toxicity can cause enteritis, diarrhea (Aggarwal *et al*., 2007).

Lead poisoning is serious health problem, it can happen if builds up in human body, over many months or years, it is harmful in adults, but is more harmful to children according to a small body, children age six and under are at highest risk, lead source in house paint, dirt, dust, and toys are usually low levels of lead, this is often called "mild" or "low-level" exposure, adult bodies remove almost all of swallowed lead, while children's bodies only remove about a third (USEPA, 2016).

Nerium oleander is an evergreen shrub elongated up to four meters in height. and belongs to the family Apocynaceae, spread in tropical Asia *Nerium oleander* is cultivated worldwide as an ornamental plant, it is native to the Mediterranean region, also found in Southern Europe and Southwest Asia, it is naturalize very easily and in many areas plants are sub-spontaneous (WCSP, 2014).

Leaves are narrow acute at the apex, elongated from 10 to 20 cm, shortly petiolate, with coriaceus dark green blade narrow, short stalked and dark or grey green color and un toothed. Some cultivars have leaves variegated with yellow or white patches, leaves have prominent, mid rib are "leathery" in texture and usually arise in groups of three from stem, flower of the plant have terminal heads, sometimes white and pink, each flower is about 5cm in diameter, five petalled, although sometimes cultivators have double flowers, branches of oleander characterized as a flexibility with green, smooth bark eventually tending to grey in mature plant (Diane *et al*., 1999).

All plant parts, involving milky, white sap are toxic, and cause adverse reaction, when uptake by living organisms, It has numerous toxic compound and most toxic components are oleandrin and the cardiac glycosides neriin (Abdou *et al*., 2019). Uptaking parts of *Nerium oleander* cause hyperkalemia, nausea, abdominal pain, vomiting, dysrhythmias, and diarrhea (Bandara *et al*., 2010). It used as rats poison, the pounded leaves used as an insecticide (Kirtikar and Bassu, 1999).

This epidemiologic reports showed that age most vulnerable to oleander exposure in Texas is young kids, especially from l-year old children. No death states from oleander uptake were recorded in Texas in 1994 (Langford and Boor, 1996). In 2000 two Russian kids were dead while they were sleeping, after autopsy was conducted by the forensic doctor, they found cause of death was poisoning by oleandrian (A.P, 2018).

Poisoning states in women was admitted into emergency room and diagnosed with symptoms of dizziness and constant vomiting in period about 15 hours from eating the oleander 10-20 leaves (Khan *et al*., 2010).

Aims of The Study:

To determine the effects of some Siqua constituents like lead and *Nerium oleander* on some organs in Male white mice. Following objectives are:

1- To investigated the histological changes in brain, small intestine, liver and kidney in Male white mice.

2- To determine the some biochemical parameters after treated with lead and *Nerium oleander*.

2.1: Siqua

Alsiqua is a treatment used in cases of severe diarrhea in children. This treatment is prescribed by people who are not doctors or have no knowledge of pharmacy matters. The components of the siqua differ from one person to another, who describe the use of siqua and are known as clairvoyants, The components of the sequoia differ from one person to another, who describe the use of sequoia and are known as clairvoyants or users of Arab medicine or herbal medicine.

The components of the siqua are a mixture of several sources, the plant source and it consists of beneficial plants and harmful plants represented by the *Nerium oleander* plant. The mineral source is lead and ink, and the last source is the animal source, which is the skin of a urchin or the nails of either animal or a human. Siqua is used by people with low understanding and no knowledge of the toxic and dangerous damage to the components of the siqua. The use of siqua causes serious and dangerous damage, for example, disruption of the digestive system, defect in nervous system, weakness of the limbs, anemia, damage in the kidneys, delaying growth, and finally death in critical cases.

2.2: *Nerium oleander*

Nerium oleander is adornment plant, that grown in public city and gardens, *Nerium oleander* leaves have a leathery and linear, colors differs from green to grey-green and yellowish veins, *Nerium oleander* flowers are fragrant and funnel-shaped, with white to pink to deep red colors. fruits of it are a narrow sheath containing silky-haired seeds, this plant distributed originally in the Mediterranean region and subtropical Asia, but now is growing in many regions of the world, like in United States, China, Australia and Middle East countries (Derwich *et al*., 2010).

Oleander is green plant, widespread round the world, belonging to the Apocynaceae family; mall tree from 1 to 10 m in tall containing sticky gummy sap in the dogbane. Oleander is an idiom for plants of the *Nerium oleander*, *Nerium odorum,* and *Nerium indicum* species. whose most prevalent species are *Nerium oleander* (common oleander),(Shepherd, 2004).

Its toxicity has been known since ancient times: in India, before Christ, the shrub was called Kajamaraka, "the herb that makes the horse die" (Ceruti *et al*., 1993).

2.2.1: Taxonomy of *Nerium oleander*

- Domain: Eukaryota

- Kingdom: Plantae
- Phylum: Spermatophyta
- Subphylum: Angiospermae
- Class: Dicotyledonae
- Order: Gentianales
- Family: Apocynaceae
- Genus: Nerium
- Species: *Nerium oleander*

Atlas of Florida Plants. Retrieved 2017-05-07.

2.2.2: Toxicity of *Nerium oleander*

All parts of the plant are highly toxic because it contain several non-digitalis cardiac glycosides, including oleandrin, digitoxigenin, nerin, andolinerin, collectively referred to as cardenolides, caused from relatively high lipophilicity resulting in a rapid and extensive gastrointestinal absorption and a slow urinary excretion level, most active molecule in plant is oleandrin (Praveen *et al*., 2012).

Children older than six years are exposed to this plant. Where one leaf could be lethal, more serious toxicity is usually resultant of suicidal attempt by adults (Bandara *et al*,. 2010).

Rubini *et al*., (2019) showed the effect of *Nerium oleander* on cows, veterinarians found dead case, samples were taken from rumen content, samples were laboratory examined, there was presence of oleander leaves confirmed.

Grazing animals can ingest parts of shrub in arid areas or during dry season when there are scarcity of fodder, poisoning may be attributed to human managerial errors like when oleander is unintentionally mowed, crushed, and mixed with feed, animals may also intoxicated after the ingestion water containing macerated leaves (Praveen *et al*., 2012).

Adam *et al*., (2001) studied oral administration of *Nerium oleander* leaves on the sheep, oral doses of 1 or 0.25 g of dried *Nerium oleander* showed ruminal bloat, dyspnea, incoordination of movements, recumbency, limb paresis, and death 4-24 hours after dosing. Lethal dose of *Nerium oleander* varies according to the animal species.

Even though cardiotoxic glycosides act at low concentrations and the intoxication is potentially fatal, there was a woman committed suicide alone in her car, parked at a rest stop of a trunk road, so that when the Police found her, she was already death, the autopsy, performed three days after the woman's death, revealed a piece of oleander leaf on the posterior third of the tongue's body and several plant residues, similar to the one recovered on the tongue, into gastric content. Petechiae on the deep multi-organ congestion, surface of the scalp, and pulmonary edema also observed (Azzalini *et al*., 2019).

Toxicological investigations performed samples collected during autopsy and aimed at detecting the active substance oleandrin, the detection of oleandrin in biological cadaveric samples revealed high, fatal, concentrations: 82.86 ng/ml in cardiac blood, 36.15 ng/ml in peripheral blood, and »100 ng/ml in gastric content (Azzalini *et al*., 2019).

In fact, blood levels of about 1–2 ng/ml of oleandrin are regarded as toxic10 and cases of fatal acute poisoning described for blood levels of about 9.8–10 ng/ml 6,11. It is interesting to refer to the case of a person who died 5 h after the ingestion of 25 oleander leaves: toxicological analysis of cadaveric samples revealed concentrations of oleandrin of about 66 ng/ml in blood and of about 254 ng/ml in urine (Baselt, 2017).

Akhtar et al., (2014) show effects of *Nerium oleander* on red blood cells, hemoglobin, white blood cells, platelets and lymphocyte led to significant variations in these parameters.

Initial management of oleander poisoning is directed to supportive care and symptomatic treatment, patients may need resuscitation depending upon severity of the poisoning, and intravenous fluids should be given to control hypo volemia due to excessive vomiting and diarrhea, while uncommon in the United States, yellow oleander ingestion is the preferred method of suicide in Sri Lanka and parts of India because it is so rapid obtainable. Clinicians practicing in these areas should be aware of the presentation of oleander toxicity due to the possibility of either purposeful or accidental ingestion (USDA, 2012).

When tissues are exposed to damaging conditions, intracellular enzymes leak from injured cells into the systemic blood circulation or may be found in urine. In general toxicology studies, changes in specific enzyme levels are one of the most common markers of target organ toxicity. The most common measured enzymes are

alanine aminotransferase, aminotransferase, aspartate creatinine, urea changes in oleander toxicity (Khordadmehr *et al*., 2017).

Many cases of poisoning animals with oleander, and then the death of animals due to the effects of poisoning, two American horses suffered from lethargy and an irregular heartbeat, after that the first horse died and the second was sacrificed in order to find out the cause and examined the animal after laboratory examination, the presence of oleander in digestive system contents of first horse and digoxin in the serum of the second horse (Butler *et al*.,2016).

A female dog ingested oleander poisoning and diagnosed with vomiting, diarrhea, and irregular heartbeat and then dogs were treated but didn't recover for unknown reasons (DeClementi *et al*., 2017).

Also, sheep were infected with oleander poisoning, and all the animals died, after animal dissection and laboratory examination, extensive bleeding observed, as well as presence of oleander leaves in the contents of the digestive system (Soto-Blanco *et al*., 2006).

In another case, a fetal poisoning with *Nerium* in a person and after analyzing blood samples, the presence of oleandrin in blood samples with a ates ~10 ng/ml (Wasfi *et al*., 2008).

2.2.3: Effects of *Nerium oleander* on organs system

Ingestion of this plant can affect on the heart, gastrointestinal system, and the central nervous system, the gastrointestinal effects can consist of nausea and vomiting, abdominal pain, excess salivation, diarrhea sometimes contain blood, especially in horses. Cardiac reactions consist of irregular heart rate, sometimes characterized by a racing heart at first that then slows to below normal further along in the reaction, extremities may become pale and cold due to poor or irregular circulation, the effect on the central nervous system may show itself in symptoms such as drowsiness, tremors or shaking of the muscles, collapse, seizures, and even coma can lead to death (Oleander, 2017).

2.2.3.1: The effects of *Nerium oleander* on brain

Ni *et al*., (2002) studied the effects of oleandrin on the brain of mice after intraperitoneal injection with (3mg/kg), the oleandrin concentration in the brain is high and oleander content enhance transport of oleandrin across blood brain barrier then showed the pathological change in the Central Nervous System of mice

involving perivascular and perineuronal oedema and hyperaemia, aiso cause with higher toxicity, multifocal haemorrhage and liquefactive necrosis were observed.

Omidi *et al*., (2011) showed that toxicity of *Nerium oleander* on the brain of male chickens cause acute congestion, necrosis in nerve cells, infiltration of inflammatory cells in the perivascular.

Majeed, (2012) studied the effect of *Nerium oleander* on the brain of Albino white mice which showed coagulative necrosis, haemorrhage, and edema, so, increasing in levels of *Nerium* extract toxicity cause severe damage.

2.2.3.2: Effects of *Nerium oleander* on the small intestine

The result of *Nerium oleander* treated orally on goats showed degenerative in small intestine as infiltration in inflammatory cells, hyperaemia and interstitial haemorrhage (Aslani *et al*., 2007).

Yahaya *et al*., (2000) showed the effect of treatment with *N. oleander* on small intestine rats caused erosions on the intestinal epithelium, damage in the submucosa and catarrhal enteritis

However, organ destruction caused from toxic content of oleander glycoside which was shown to act on gastrointestinal tract causing enteritis, abdominal pain and diarrhea (Radostitis *et al*., 2000). The generation of free radicals called reactive oxygen species (ROS) which cause more cellular destruction, while the effects of *Nerium oleander* leaf aqueous extract on intestine cause severe haemorrhage on the serosal surface, haemorrhages were present in the mucosal surface, necrosis in cells epithelia and lymphocytes were infiltrated in mucous membrane and submucosa, the histopathological result of the *Nerium oleander* effects cause acute epithelial necrosis in the proventriculus and scattered necrosis of surface enterocytes are probably direct effects of the toxins on the vascular endothelial bed (Aslani *et al*., 2007).

Reznick *et al*., (2006) showed the effect of treatment with *N. oleander* on small intestine mice caused haemorrhagic, degenerative and necrotic changes and the eosinophils were infiltrated in mucosal and submucosal layers of this organ.

2.2.3.3: Effects of *Nerium oleander* on liver

The biochemical effects of *Nerium oleander* toxicity on the liver showed change in alanine aminotransferase (ALT), aspartate aminotransferase (AST) due to liver injuries, when the liver of male albino mice exposed to *Nerium oleander* leaf extract cause the evaluation the level of this enzyme (Narayane *et al*., 2009).

Jaafar *et al*., (2019) noted effects of Nerium oleander on liver led to dilatation in central vein, enlarged vaculative hepatocytes, fibrosis perivascular perihyperplastic bile duct, hyperplasia of bile duct and thickening of blood vessels wall.

In a study of the effect of *Nerium oleander* in the liver, there was mononuclear cell infiltration in portal spaces. Also, noted congestion and hemorrhage in some area, individual cell necrosis and dilation of sinusoidal spaces (Omidi *et al*., 2011).

Al-Hakak *et al*., (2019) has shown that section of liver mice treated with alcoholic extract of *N. oleander* showed programmed death in hepatocyte, enlargement of the germs, cell membrane thickness, emergence of hemosiderin also accumulation of fats.

Changes happened in the liver when absorption of *Nerium oleander* and caused early analysis cellular to some areas, necrosis in walls of blood vessels and damaging in some of bile ducts, existence of decomposing in acidic cells and liver plate necrosis (Narayane *et al*., 2009). When rabbits were extracted with crude alcohol extract of the seeds noting that proliferation of hepatic cell, toxicity of the extract leads to an imbalance in the effectiveness of liver enzymes. If toxin removed, toxic matters are associated with liver cells so cause necrosis, then causing liver damage and cell death (Mohammed and Abdullah., 2002).

2.2.3.4: Effects of *Nerium oleander* on the kidney

The results of goats that treated orally to *Nerium oleander* showed histopathological changes like degeneration in bowman capsule, shrinkage of glomeruli, dilatation in bowman capsules, and renal necrosis at convoluted and collector tubule, and necrosis of tubular epithelium (Barbosa *et al*., 2008). Dried *Nerium oleander* leaves effected on kidney widespread tubular epithelial necrosis in distal and proximal tubules, congestion vascular and necrosis in collecting ducts (Aslani *et al*., 2004).

Khordadmehr *et al*., (2017) observed clear histological changes in kidney resulted from toxic effects of *Nerium oleander* which cause hemorrhage, inflammatory cell infiltration, hyperemia, and interstitial nephritis.

Histological changes in rats kidney that treated to *Nerium oleander* which digoxin cause vacuolation of the urinary tubules epithelia and vacuolation of urinary tubules enlargement of Bowman's space (Jaafar *et al*., 2019).

The level of urea serum and creatinine serum in wistar rat and mice after treated with *Nerium oleander* extract cause increase in levels of these enzymes (Khordadmehr *et al*., 2017).

2.3: Lead

Lead is a heavy metal that exist as an oxide or a salt or, it is one of harmful and dangerous environmental pollutants (Milens *et al*., 2006). Source of lead from environmental and industrial exposure and cause public health problems, lead poisoning cause a variety symptoms, signs that appeared on human vary depending on individuals and period of lead exposure (Karri *et al*., 2008). Symptoms are nonspecific and it may be subtle, but may some people with high lead levels and have no symptoms (Kosnett, 2007).

According to WHO, (2016). lead is caused about 540,000 deaths round the world, it happen most spread in the developing countries, Those who are in larger hazard, and its reckon to result 0.6% of world's disease onus (Needleman, 2004).

The World Health Organization published list of 10 chemicals impact on human health included lead (WHO, 2016).

Ekong *et al.,* (2006) mentioned that lead is an important element for industry, but it has no useful function in human body.

Institute for Health Metrics and Evaluation (IHME) presented that people exposed to lead accounted 1.06 million deaths added to 24.4 million years of healthy life absent (disability-adjusted life years (DALYs) worldwide according to prolonged-period effects on health, greater charge was in low- and moderate countries. Also IHME published that in 2016, exposed to lead accounted for 63.2% of universal onus of unknown reason developmental mind hindrance (IHME, 2017).

Symptoms continued over weeks to months as lead accumulation in tissue caused from chronic exposure, but severe symptoms from brief, intense, exposures also happened (Mycyk *et al.,* 2005). Lead as an organic is considered more toxic compared with inorganic lead because of rapid solubility in lipid. Poisoning with organic lead compounds influenced on central nervous system, such as cognitive deficits, insomnia, delirium, tremor, and convulsions (Grant, 2009).

Toxicity determined by exposure period and lead amount in tissues and blood, cute poisoning may be from highly exposure for many time, chronic toxicity frequented few -grade exposure time by time (Pearson and Schonfeld, 2003).

After uptaking lead, its distributed in organisms by proteins or red blood cells, when lead being inside cells, most of it bounds with hemoglobin more than bounds with membranes of red blood cells, hematopoietic system is one of the most sensitive systems, blood represents not only mode of transportation, but added to the crucial toxicity target of lead (ATSDR, 2007).

When lead is orally administrated to the treated mice toxicity effects of lead was significantly decreased in count of red blood cells, also in white blood cells, lymphocytes, hemoglobin and platelets (Flora *et al.,* 2012).

Lead diagnosis and therapy depends on quantity of lead in blood, unit used to measure is micrograms per deciliter in blood (µg/dL). Levels of lead in urine can also be used, but it is not common. In case of the chronic exposure, lead measured from the maximum concentration first in the bones, next in the kidneys (Trevor *et al*., 2007).

Pollution air is another way of lead exposure, added to dust, food, and water, or consumer products, children are more effected for lead hazard (WHO, 2016). Exposure to lead in work is commonly reason of toxicity in young with appointed career at special impact (Gracia and Snodgrass, 2007). Diagnosis commonly made by gauge of the blood lead level. The Centers for Disease Control (US) demonstrated the uppermost frontier of blood lead for adults to 10 µg/dl (10 µg/100 g) but for kids 5 µg/dl, high levels of lead could detect by alteration in red blood cells or bushy lines of bones of kids then seen by X-ray.(Dapul and Laraque, 2014).

Lead exposure made several syndromes such as unnatural demeanour that differs from person to person, moreover period of exposure plays an important function (kosnett, 2007).

Generality pharmaceutical companies put extreme for ultimate for a day consumption of lead as (1.0 µg/g), long time ingestion of minimum rates of lead is dangerous to humaneness existence, rise in blood level are combined to late majority in girls, but there is no onset rate for the level currently beneath which its focus could be estimation as safe (Schoeters *et al.,* 2008).

Levels of lead in blood 25 and 60 µg/dL lead to cause nervous psychological effects such as late reaction time, irritability, onerousness in focus, also be late motor nerve connection and anemia in addition to head pain, when manifest levels of lead in blood maximum than 50 µg/dL (Merill *et al*., 2007).

When blood lead levels rise override 100 µg/dL lead to very intense symptoms, such as neuropsychiatric (brain tumor) combined with rise in pressure within hallucination ,cranium, stupefaction, and head pain, while this symptoms exhibited in children when blood lead levels more than70 µg/dL (Henritig, 2006).

Lead toxins came from uptake by drink water and food polluted with lead, episodic ingestion of polluted soil, dust could be cause poisoning, lead rapidly absorbed by blood then cause harmful effects to organs and tissues such as central nervous system, and kidneys cardiovascular (Bergeson, 2008).

Lead exposure results coma, anemia, and death (ATSDR, 2007). Not obvious lead toxic with signs abdominal , ache, and stomach pain and those could not be diagnosed and perhaps unnecessary intestine eleminate and abdominal surgery, patient with lead toxicity suffered from acute abdominal pain also hemolytic anemia that beaded not specific diagnosed of lead toxic. Chemical factors at crucial time of accumulative lead is changing histology and physiology of systems in organism (Sharma *et al*., 2013).

Correlation between oxidative stress and lead toxicity, focused on matters which have antioxidant features for define effects against it caused toxic lead effects mammalian organs by less antioxidant reverse, and generating reactive oxygen species. ROS cause a change in cellular membrane and tissue (Dewanjee *et al*., 2013). Lead toxicity in systems of mammalian generates (ROS) and antioxidant, and cause vascular damage and neurololgical, Lead cuts enzyme activation, then block trace mineral absorption and binds to sulfhydryl protein, altering calcium balance (Ercal *et al*., 2001). Oxidative stress was studied in low levels of lead exposure Uruguayan children, proposition possible harmful effects of it on oxidative stress (Roy *et al*., 2015). According to Flora *et al*., 2012 oxidative stress means equilibrium production of free radicals and the biological system to capacity removal poisoning interactive medium and reform resulting wrong. Gui *et al*., (2017) studied dolphins which exposed to lead, and showed that the amount of lead that measured in the liver and kidneys was a high in accumulation of it in tissues of dolphins organs.

2.3.1: Lead poisoning is two types

2.3.1.1: Acute poisoning

In acute poisoning, neurological signs showed muscle pain, numbness, weakness, and tingling, with rarely, nausea, symptoms combined with inflammation in the

brain, diarrhea, abdominal pain, vomiting, hypertension and renal dysfunction (Martin & Griswold, 2009).

Gastrointestinal problems, represented by poor appetite, constipation, or losing weight, are widespread in case of acute poisoning. Absorption of big amounts of lead during a short time causing a shock "insufficient fluid in the circulatory system" according to decreasing water from gastrointestinal tract. Hemolysis (rupturing in red blood cells) resulting from acute poisoning, this can cause hemoglobin in urine and anemia (Henretig, 2006).

In studying acute toxicity of lead in oral administration, the results of toxicity led to significant changes in physiological, biochemical parameters of blood in mice (Yuan *et al*., 2014).

2.3.1.2: Chronic poisoning

Chronic poisoning sometimes presents symptoms which infect multiple systems, that is combined with three impotant types of symptoms: neuromuscular, neurological, and gastrointestinal (Martin & Griswold, 2009). Neuromuscular symptoms result from dense exposure, while gastrointestinal symptoms resulted from exposure to longer periods. Signs of chronic exposure are lost short term memory and concentration, depression, abdominal pain, loss of coordination, numbness, nausea, and tingling in extremities (Merrill *et al*., 2007). Sleep problems with headaches, stupor, slurred speech, and anemia also in chronic lead poisoning, case of chronic exposure lead frequently isolation of maximum concentration initial in bones then kidneys, according to US center of Disease Control and Prevention and World Health Organization, blood lead level of 10 μg/dL or upper could cause risk, lead exposure also cause weakness in growth and harmful effects even in below levels (Roosi, 2008).Burton line is blue line found along gum with black bluish edging to the teeth, this is consider as another indication for chronic lead poisoning. Children with chronic poisoning perhaps refuse playing or having hyperkinetic and aggressive behavior disorders, visual disturbance also could present with progressing blurred vision as a result of central scotoma, this caused from toxic optic neuritis (El Safoury *et al*., 2009).

2.3.2: The effects of lead on the organs system

The uptake, distribution, and finally accumulation of elements in tissues and organs depend on some factors, like the form of the element and characteristics,

dose, route, and exposure periods, the affinity of binding to ligands in cells, and sensitivity of species, hematopoietic system is one of the most sensitive organs to toxicity, in case of oral administration, lead go to intestinal and absorption then transported with blood, they can be distributed by red blood cells (Omobowale *et al*., 2014).

Half-life of lead in tissues causes in its induction to blood stream long after the initial exposure (Patrick, 2006). Blood lead had a lower half-life reached about 40 days in human, but increases in case of pregnant women, and in children with developing stage of bones, developing bones of children which undergo remodeling permits lead to be continuously reintroduced to the stream of blood (Barbosa *et al*., 2005).

Due to long time of exposure to lead reached for a years, a much slower clearance occurred, that is from prolonged accumulation in bones over a long time, along with teeth , bones, and blood ،many other tissues keep lead in the body, ، spleen, brain, liver, kidneys and lungs (Dart *et al*., 2004).

Lead effects on all body systems and organs, especially nervous system, but also kidneys, bones and teeth, cardiovascular, reproductive and immune systems (UNICEF, 2020).

2.3.2.1: The effects of lead on the brain

Nervous system is a very sensitive and lead ease to target for lead induced toxicity compared with other organ system, both central nervous system and peripheral nervous system become danger affected by lead poisining in children, while in adults central nervous system is affected, and peripheral nervous system also severely affected (Grant, 2009).

The poisoning interferes with normal children brains during development, neurochemical development (including of neurotransmitters), synapse forming in nervous system and cerebral cortex; therefore children are in more risk of lead neurotoxicity than adults (Lidsky and Schneider, 2003).

Xu *et al*., (2009) exposed 24 adult rats to 0.2% lead acetate and found that lead is able to pass through endothelial cells of blood brain barrier because it can substitute for calcium ions then up taken with calcium-ATPase pumps. It causes destroy of neurons myelin sheaths, and reduces the number of neurons; interferes with neurotransmission, and decreases neuronal growing (Guidotti and Ragain, 2007).

Lead encephalopathy is a medical for emergency causes permanent brain damage in about 70–80% of children, even those who received best relief, mortality rate of people that developed cerebral involvement is about 25%, and those who survived had lead encephalopathy symptoms by time chelation therapy was begun, in about 40% who have permanent neurological problems like cerebral palsy (Kosnett, 2007).

In young children and features who are highly vulnerable to neurological effects of lead as the developing of nervous system absorbs highly amounts of lead, the proportion of system circulating of lead and absorbing access to brain of young children is much more compared with adults, in children, higher levels of lead may be severely affected with delayed growing, losing hearing and short term memory, at extreme levels may cause brain damage then death (Guidotti and Ragain, 2007).

Repercussions of exposure to lead on peripheral nervous system had been noted in the form of peripheral neuropathy in adults, including motor activity reduction resulted from the loss of myelin sheath that envelope nerves; severely causing weakness, lack of muscular coordination especially of exterior muscles, impairing the transduction of nerve impulses, and fatigues (Devi *et al*., 2005). Within brain, lead induced destruction in prefrontal cerebral cortex, hippocampus; cerebellum can cause a variety of neurological disorders, like damage of brain, mental retardation, behavioral troubles, damage in nerve, and perhaps Alzheimer's disease, schizophrenia and Parkinson's disease (Sanders *et al*., 2009).

2.3.2.2: The effects of lead on the small intestine

One of most important systems that affected by lead is gastrointestinal system because ingested of lead is directly absorbed in it, it causes significant variations in duodenal cell proliferation and differentiation in development thud result and found after exposed albino mice (Sharma and Barber, 2012).

Lead toxicity can manifest with gastrointestinal effects, exposure to lead induces alteration in basic precursors of developing gastrointestinal tract that turn interfere with absorption of food in premature stages, increased lead levels effect on smooth muscle of gastrointestinal tract, producing a vague abdominal syndrome, that is manifested by anorexia, cramping, nausea, and a metallic taste in mouth (Sharma and Barber, 2012).

Mobarak and Sharaf, (2011) exposed fish (*Poecilia latippina*) to lead acetate which caused irregularities in microvilli cells and hypertrophy, fusion in intestinal microvilli and necrosis.

Sharma *et al*., (2013) treated adult Swiss female mice with lead acetate, this treated caused degeneration changes in submucosa and hyperplasia in squamous mucosa, shrinkage in squamous mucosa and ertrophy in keratinocytes, Disappearance in keratinized layer of squamous and Muscularis externa was necrotic.

Abdou, and Hassan, (2014) Treated rats once a day by oral gavage during five days with lead acetate 200 mg/kg BW, results showed that lead treatment could induce mucosal damages of stomach, infiltration of inflammatory cells into the lamina propria and villus damage of small intestine.

Crypts architecture was largely destroyed and submucosal layer was thickened compared with control and components in both longitudinal and circular layers of muscularis externa were not identified, and observed degenerative in submucosa and distorted muscularis externa in stomach in lead (Sharma *et al*., 2013).

Necrosis causing cell death in columnar epithelial cells at the tips of villi in the anterior intestine, necrosis and shortening of villi in posterior intestine, Since gastrointestinal mucosa is the first target organ to lead and intestinal inflammatory cells are charged for providing protect against pathological damage caused from toxicity, lead exposed group and columnar epithelium that covered villi was not clearly seen and epithelial cells were intermingled (Tomczok *et al*., 1988).

It was suggested that lead increases formation gastric ulcers when interfers with the oxidative metabolism in the stomach that increased the incidence of gastric ulcer).

The implication of lead is causing increase in forming free radicals, that if not mopped up by free radical scavengers, expose the stomach to inflammation with gastric mucosal damage (Olaleye *et al*., 2007).

2.3.2.3: The effects of lead on the liver

Liver plays an important role in detoxification processes, function of it affected by lead toxicity (Haleagrahara *et al*., 2010). Also liver is the largest store of lead between soft tissues following by kidneys (Mehana *et al*., 2012).

Lead are transported to liver, it can cause damage and defect in function, liver damage could be confirmed by histopathological findings, and is often accompanied by increasing blood enzyme rates, reducing protein synthesis, toxic effects on kidney are represented by the structure damage of kidney and abnormal in excretory function *(*Abdou and Hassan, 2014).

Abdou and Hassan, (2014) studied lead acetate by injection wistar albino female rats, the study showed that dilatation of the blood sinusoids, degenerated hepatocytes and pyknotic nuclei, vacuolated cytoplasm, lymphocytes aggregation inside hepatic tissues and loss cellular architecture.

Lead hepatotoxicity in Wistar (type of rats) had been elevation in the levels of serum liver enzymes alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) and alterations in the hepatic cholesterol metabolism (Omobowale *et al*., 2014).

Liver is main target of lead that induced oxidative stress; acute intra peritoneal administration 15mg lead acetate/kg for 7 days showed inflammation and necrosis in liver parenchyma (Mohammadi *et al*., 2014).

Daniel (2013) treated twenty male rats with lead (250 mg/kg) and blood samples were collected and serum of liver enzymes analyzed, accumulation of amounts of lead were in oxidative stress in the liver tissue.

Nwanebu (2019) showed histopathological alteration in the liver tissue of Wistar Albino Rats when treated with lead acetate, the effect of lead was obvious and deleterious on the liver, showed destruction with irregular hepatocytes, inflammatory cell in hepatic tissue, congestion, mononuclear infiltration in the portal triad was seen, and dilated of central vein and sinusoid.

The liver of rats after expose to lead caused necrosis to the hepatocytes, mild fibrosis, fatty changes, hepatocyte proliferation, hydropic degeneration and, biliary hyperplasia (Mehana *et al*., 2012).

Lead acetate caused injury to the liver of rats by histopathological changes, induced hepatotoxicity and rise total bilirubin, alkaline phosphatase and γ-glutamyl transpeptidase (Moneim *et al.*, 2011*).*

Hegazy and Fouad, (2014) studied the effects of lead on the liver of rats by giving 0.13% lead acetate solution in drinking water for 4 weeks, results showed degenerative of cytoplasm, inflammatory cells near central vein hepatic cell, necrosis and infiltration.

2.3.2.4: Effects of lead on the kidney

Kidney damage occurs when exposure to high rates of lead, and the evidences suggest that lower rates can cause damage to the kidneys and causes nephropathy that may cause Fanconi syndrome, in this case the proximal tubular function of kidney is impaired (ATSDR,, 2007).

Long term exposure to levels lower could cause lead nephropathy as nephrotoxic in patients from developed countries which had chronic kidney disease and were at risk because of diabetes mellitus or hypertension (Rubin and Strayer, 2008).

Lin and Huang, (2003) was checked patients which had abnormal serum creatinine levels, they found that lead stored in the body blocked the excretion of urine and cause accumulation of urea and waste products in the body, especially in patients who had renal insufficiency.

Lead induced renal dysfunction could be classified as acute and chronic nephropathy, and occurs mostly at high rates of lead exposure >60 μg/dL in blood but damage at lower rates had been reported ~10 μg/dL blood (Grant, 2009). Lead nephrotoxicity induced progressive tubular and glomerular alterations like disruption Bowman's capsule, destruction of epithelium lining the tubules, shrunken glomeruli with capsular space, and dilation in renal tubules, blood congestion and swelling of convoluted tubules (Nwanebu, 2019).

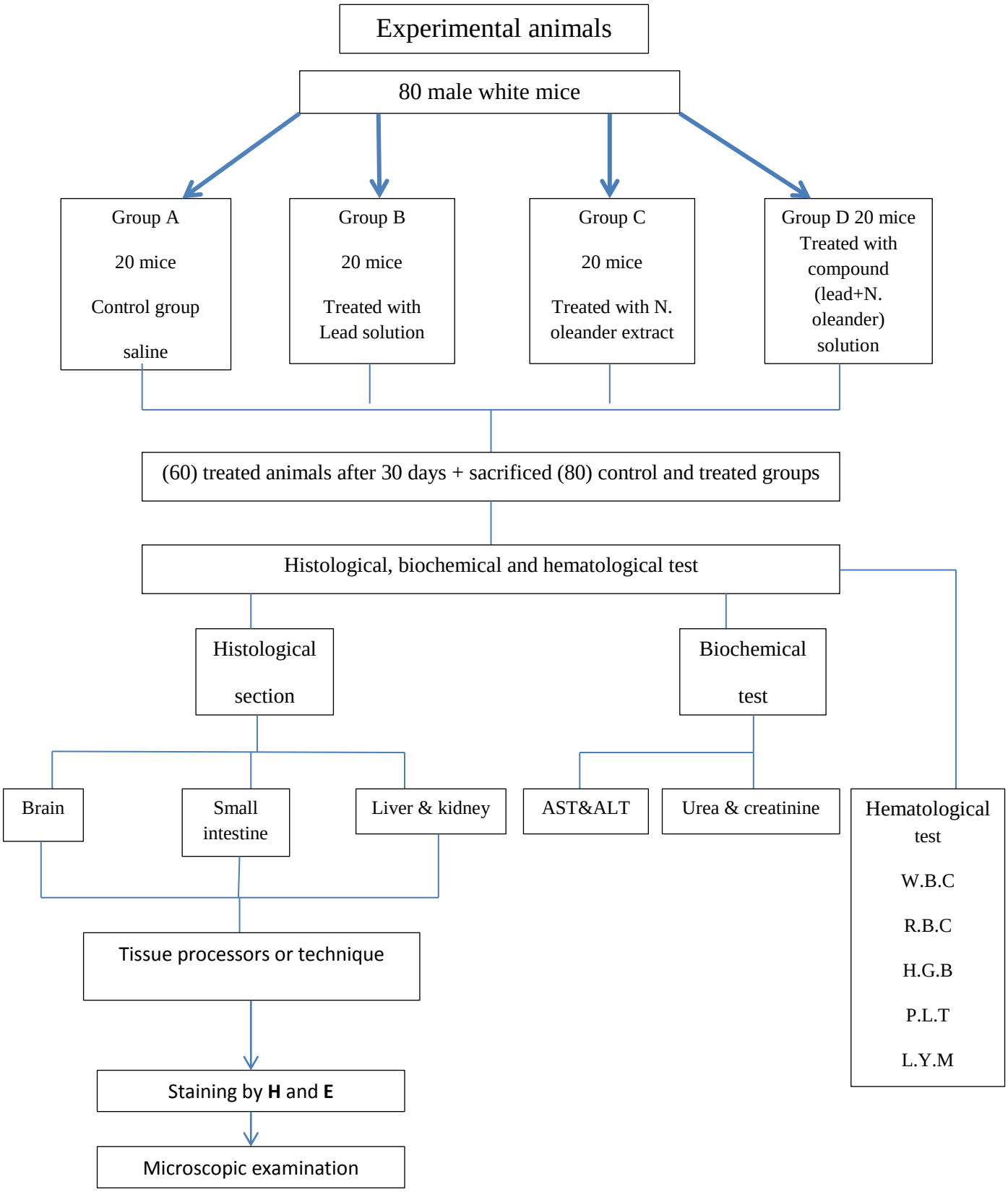
Sujatha *et al*., (2011) had shown the section of the kidney after treated by lead for 12 weeks showed congested blood vessels, hyperplasia of tubular epithelium, intra tubular hemorrhages, desquamated tubular epithelial cells, and increased of apoptotic bodies in the proximal convoluted tubules.

Chronic lead nephropathy occurred from years of lead exposure that manifested in kidney biopsy, test of serum creatinine, the result of biopsy was mild focal leanness, and prominent interstitial (Benjelloun *et al*., 2007).

The accumulation of reactive oxygen species in kidney tissue cause renal dysfunction, Bowman's spaces were existed, cellular damage and tubules desquamated cells, (Matović *et al*., 2015).

3. Materials and Methods

3.1 Experimental Design



3.2 Materials

3.2.1 Experimental Animals

The eighty animals were divided into four groups (A, B, C and D), each group was composed of twenty male mice.

Group A which consider as a control group, the group B was treated with lead solution, group C treated with *Nerium oleander* and group D treated with mixed lead and *Nerium oleander*, the treated groups were treated by main compositions of siqua for thirty days.

The experimental animals living in laboratory plastic cages, all cages put in animal house of the science of college in Al Muthanna University. The main weight of experimental animals was (30 gm), the animal house temperature was (25-28) ºC and humidity about 40 to 45%, the animal feeding with standard pellets and normal saline. The preparation and treatment of the animals study take up from 1\ 1\ 2019 until 1\ 3\ 2020, the treatment animals with the main compositions of siqua continue for thirty days only from 1\ 2 \ 2020 until 1\ 3 \ 2020.

3.2.2: Chemicals:

Table 3-1: Chemicals with Their Productions

No.	Chemicals	Productions
1	Formalin 37 %	Linear chemicals, Barcelona-Spain
2	Chloroform	India
3	Glycerin	England
4	Paraffin wax	England
5	Hematoxylin crystals	England
6	Eosin	England
7	Oil emersion	China
8	Absolute Alcohol	Japan
9	Xylene	England

Table.3.2: Instruments with their Productions.

No.	Tools	Productions
1	Filter paper	China
2	Disposable surgical Gloves	Malaysia
3	Cotton	Jordon
4	Refrigerator	Concord
5	Warring Blender	Crown
6	Gel tube	Celeco- Jordon
7	EDTA tube	China
8	Slides	China
9	Cover Slide	China
10	Ependroff tubes	Germany
11	Beakers	China
12	Disposable syringe	China
13	Sterile bottle	Jordan
14	Petri dishes	China
15	Electrical balance	Shimadu company- Japan
16	Centrifuge	GMBH- Germany
17	Microscope camera	OMAX-China
18	Microtome	Mycrom-Germany
19	Oven	Memmert- Germany
20	Cobas c 111	Germany
21	Light compound Microscope	GENEX-
22	Micropipette	Huawei –China
23	Water path	Germany
24	Surgical set	England
25	Processer	Cyan-Germany
26	Bluking	Thermo-England

27	Container	China

Table.3.3: The stains

Hematoxylin and eosin stain	1.0 gm
Distilled water	1000 ml
Sodium iodate	0.2 gm
Ammonium or potassium alum	50.0 gm
Citric acid	1.0 gm
Chloral hydrate	50.0gm

Table 3-4: The Biochemical Kits and Their Production

No	Test Kits	Productions
1	AST	Germany
2	ALT	Germany
3	Urea	Germany
4	Creatinine	Germany
5	C.B.P.	China

3.3: Methods

3.3.1: Plant material:

3.3.1.1: plant collection:

Nerium oleander leaves collected from markets in Al- Samawa city. The leaves cleaned and dried at room temperature then crushed by a blender at the same day of preparation of the extract.

2.2.1.2: preparation of aqueous extract of *Nerium oleander* :

According to (Hernandez *at el*., 1994), mode (10 gm) of plant powdered added to (200ml) of distilled water in a sterile glass beaker and left 24 hours with continued jolt ,then passed on the layers of sterile soft cloth for its candidacy and then separation of the filterate by using a centrifuge (3000/rpm) and then dried the liquid in an electric oven degree (40 c) for drying the extract. Then collected and placed in a sterile bottle and preserved in refrigerator to until use.

3.3.1.3 Concentration prepares of *Nerium oleander* (stock) is:

Dissolving (500) mg active ingredient (*Nerium oleander*) in (50) ml DW(Salih, 2008).

Mean: (500) mg /(50) ml (DW) → (500,000µg /50) ml DW = (10,000)µg /ml.

Mean: each (0.1) ml of stock solution equal to(1000)µg active ingredient.

Dose (250)mg/kg convert to µg/gm. equal (250) mg/kg.

Example: mouse weight = (30) gm. →(250) µg ×(30) gm. =(7500) µg

Mean: mouse dosing is about (0.75) ml.

3.3.1.4 Concentration prepares of lead (stock) is:

Acute LD50 of Lead was (60) mg/kg when administered orally to mice (Jawad and Bassim, 2012).

Dissolving (30) mg active ingredient (lead) in (10) ml DW.

Mean: (3) mg / ml DW → (3000)µg /ml DW.

Mean: each (0.1) ml of stock solution equal to (300) µg of active ingredient.

Dosage : (30) µg/gm.

mouse weight = (30) gm.→ (30)µg × (30) gm. = (900)µg.

Mean: mouse dosing is about (0.3)ml.

3.3.2 Administration of experimental animals:

The animals were administrated with lead (0.3)ml and aqueous extract of *Nerium oleander* (0.75) ml to period 30 days

Group (B) were administered with 0.3 ml of lead .

Group (C) were administered with 0.75 ml of extract of *Nerium oleander* .

Group (D) were administered with 0.3 ml of lead and 0.75 extract of *Nerium oleander* .

3.4. Collecting blood and histological samples:

3.4.1 Blood samples collection:

The mice were anesthesia after the administration after each 30 days from administration period; three milliliter of blood was attained by cardiac puncture and transferred to gel tubes and to EDTA anticoagulant tubes to examine hematological test. The blood for the complete test is placed in the EDTA tubes, after which the blood components are measured in the Cobas c 111.

The other samples blood was left at room temperature for 15 minutes after centrifuged with (3000 rpm) for 15 minutes the serum was collected ependroff tubes and divided into equal amount (50µl) and frozen until use for laboratory assessments, to purpose examine AST, ALT, urea and creatinine.

3.4.2 Histological technique:

The tissue samples taken with $1cm^3$ after sacrificed of experimental animals and taken the brain, small intestine, liver, kidney. The tissue samples were passed through the histological technique which including many stages.

1- Washing: The tissue samples are cleaned with tap water for 1 hour after fixation to remove unnecessary fixatives from the tissue to avoid intervention with future operations. Wash is also performed with running water. (Vacca,1985; Luna,1968).

2- Fixation: The tissue samples were put in a labeled container filled with 10% formalin for 48 hours, and shaky the container gently several times, to ensure that the fluid reached all

surface, and that pieces were not sticking to the bottom or sides (A shank of glass wool placed in the container will aid in keeping the tissue free of the bottom).

3- Dehydration: The samples were taken to clear all extractable water by dehydrated diffusing through the tissue, alcohol was used. The dehydration process proceeded by increasing the volume of alcohol from 50% to absolute alcohol (50%, 70%, 80%, 90%1, 90%2, 100%1, 100%2).

4- Clearing: this stage is considered as a middle step, it is obligatory because the used alcohol for dehydration will not dissolve or mix with molten paraffin. Thus tissue must be submerged in some fluid miscible with both alcohol and paraffin before infiltration can take place. Clearing was too removed or clear opacity from dehydration tissues and making them transparent. Therefore, blocks of tissue seemed almost crystalline and never milky. Xylene was reagents used for clearing (Luna, 1968).

5. Embedding (Blocking): Paraffin wax is considered to be either soft (melting point 50-52̊ c) or hard (melting point 60-68 c). Hot weather will power the use of harder paraffin. Paraffin must be fully molten to infiltrate the tissue effectively. Embedding with paraffin. Paraffin was permitted to solidify around and within the tissue. The tissue was put in a small container or a paper box is already filled with malted paraffin. Orientation of the tissue is important in order to limit the proper surface for sectioning (Edward, 1962).

6. Cutting: Cutting was done by using the rotary microtome. The thickness is (5 µm). The samples were put in hot water path with (52̊ c) for extending tissue, then the samples were carried on the slides which had a thin layer of Mayer egg albumin, the slide was put on the hot plate with (40̊ c) for overnight.

7. Staining: (H & E) stain: For the demonstration of the general structure of the tissue (Luna, 1968)

3.7 Statistical analysis:

Experimental results were analyzed by used complete random design (C.R.D) in one way (ANOVA) to analyzed using (SPSS) (SPSS, 2012), to compare between the control and treated mice groups results, we used Duncan test for multiple range (Duncan, 1955), Data were expressed as mean ± standard deviation, the differences were considered to be significant when P value was <0.01.

4.1: The Histological Results

4.1.1: The Histological Results of Brain

4.1.1.1: The Histological Results of Control Group

The histological results that obtained from control group showed that the histological structures of brain was devoid of any histological changes. The cerebral cortex was distinguished by its normal number of tissue layers, which were differentiated into six layers, each layer differing from the other in the shape of the cells that make up each layer (Fig.4-1).

The cellular component of brain included nerves and glial cells have normal distributed among tissue layers of cerebral cortex (Fig.4-1,2).

The cerebral cortex arrangement as six layer of cellular structure, the first layer called molecular layer was composed of nerve cells arranged horizontally, the second called layer external granular layer was relatively thin layer composed of numerous small, densely packed neurons, the third called layer pyramidal layer was composed of medium-sized pyramidal nerve cells, the fourth called layer was inner granular layer was composed small, irregularly shaped nerve cells, the fifth called layer ganglionic layer was composed large pyramidal cells, the last called layer multiform layer was composed polymorphic nerve cells (Fig.4-3).

The cortex of cerebellum was arrangement in three layer of cellular structure, the first layer called molecular layer which composed of two types of cells, stellate cells and basket cells, the second layer called Purkinje cells layer was composed of large Purkinje cells, the last layer called granular layer was composed of densely populated by small granular cells (Fig.4-4).

4.1.1.2: The Histological Results of Brain Treated Group with *Nerium oleander* after 30 days orally administration

The result of brain tissue section in treated mice with *Nerium oleander* have acute histological changes, the tissue section showed glial cells hypertrophy in the peripheral zone of brain, wide empty space in the gray mater these space referred to acute degeneration in the cellular compossion of brain.

The histological results of brain noted the glial cells have large spherical nuclei with dispersed chromatic materials, on the other hand the most nerve cells in the gray matter have cell with abnormal nuclei and pale in color and heterogeneous chromatic

material (Fig.4-5) these result may be because gray matter destruction by *Nerium oleander* lead to changed in the shape of nuclei and degenerative of cellular component, the result confirmed with (Aslani *et al*., 2004) which noted *Nerium oleander* caused degeneration in gray matter tissue.

The tissue section of brain showed prominent blood hemorrhage and blood congestion in different location in trabeculai or septa, abnormal cellular aggregation around the blood congestion in the trabiculai (Fig.4-6). This result of tissue section of brain may be because high toxicity of *Nerium oleander* which lead to cellular destruction of were these results similar to (Khordadmehr and Nazifi, 2018) which noted the brain of mice when treated with *Nerium oleander* caused hemorrhage in brain.

The tissue sections of brain showed prominent histological destruction and necrosis in the brain tissue, so, noted nodular formation in irregular shape in different locations of brain, the tissue section showed abnormal cells have oval or spherical nuclei nearly from the nodular lesions, the other cells have large spherical nuclei with granular chromatic material (Fig4-7). These histological changes may be because of the *Nerium oleander* contain oleandrin which penetrated the blood brain barrier which lead to cellular destruction and changes in structures of barrier these result agreement with (Ni *et al*., 2002) which showed oleandrin caused damage in nodular formation and degenerative in nuclei cells.

The tissue section of brain after treated with watery extract of *Nerium oleander* showed prominent cytoplasmic vacuoles in the nerve cells, the results appeared many spots of Apoptosis seen in the different regions of gray matter in the cerebrum of brain, so, the tissue field showed irregular elongated space in the peripheral margin of brain, most glial cells exactly the astrocyte have dark oval nuclei (Fig.4-8). These histological changes may be because of the high oxidative stress of *Nrium oleander* caused Apoptotic cells and damage in gray matter of brain result agreement with (Khaleel *et al*., 2019) which noted the oxidative stress caused Apoptosis spots and degenerative of brain tissue.

After treated with *Nerium oleander* the brain have abnormal cells, the tissue section showed the gray matter in the cerebrum of brain have large cells, irregular in shape, acidophilic, cytoplasm and prominent dark nuclei which surrounded the empty space and aggregation in the peripheral regions of cerebrum, the results showed glial cells that aggregation between the nerve cells with large dark oval

nuclei while some of glial cells lost their nuclei (Fig.4-9) these histological changes may be due to treated with *Nerium oleander* caused glial cells proliferation that lead to alteration in shape of nerve cells, these result similar to (Aslani *et al*., 2004) which noted injury varying in nerve cells, oedma and direct effect of the toxins on the cells.

The histological results noted cellular degenerative which lead to different spaces contained the residual structures of cellular destruction, which appeared as irregular cellular spots , these findings were clearly noted in the medial regions of gray matter of brain, these tissue fields were abnormal compared with brain in control group (Fig.4-7).

The tissue section of brain of treated animal showed wide dark irregular spots have high cellular proliferation, these dark spots were surrounded by blood congestion and necrosis lesions, in some regions noted elongated space filled with blood, generally in these tissue field appeared high blood congestion in the peripherally in the meninges layer, these results may be due to the destruction or acute degeneration in the peripheral regions that covering by the pia matter, which lead to invasion into blood in the peripheral regions of brain (Fig.4-10) these histological results may be high concentration of *Nerium oleander* lead to necrosis and blood congestion these result agreement with (Khaleel *et al*., 2019) which noted the oleandrin caused haemorrhagic and necrotic lesions in the gray matter and peripheral regions of brain.

4.1.1.3: The Histological Results of Brain Treated Group with Lead after 30 days orally administration

The tissue findings of brain after treated with lead solution noted the glial cells have prominent dark nuclei, the glial cells were aggregation as small clusters in different regions of brain (Fig.4-11). This result may be due to lead toxicity which caused damage in brain tissue. These histological change of brain constant with (Manal *et al*., 2010) which noted inflammatory cell infiltration and neural degeneration.

The histological results of brain appeared prominent wide vacuoles in many location of brain, with acute degeneration, prominent lesions of necrosis showed in brain with dark spots that located in the cystic dilation in peripheral regions of brain, some of the spaces filled with blood and showed abnormal cellular proliferation a rounded the spaces, the tissue section showed progressive tissue of brain inside the spaces (Fig.4-12). These results may be due to effects by lead, this result agreement

with (Manal *et al*., 2010) which noted the brain when treated with minerals that caused neurodegenerative effects on the histology of brain tissue.

The tissue section of brain showed high cellular proliferation of glial cells peripherally of gray matter, the results showed elongated space under the blood congestion, the tissue results showed prominent blood hemorrhage of brain tissue which appeared as irregular elongated space filled with blood, isolated pia mater layer from brain tissue superficially (Fig.4-13), these histological results may be due to accumulation of lead which lead to damage in nerve tissue, this result similar to (Zahroon, 2009) which noted the brain of mice when treated with lead caused hemorrhage and destruction in brain tissue.

The tissue section of brain after treated with lead noted abnormal arrangement the nerve cells in gray matter of brain, some of cells appeared as irregular aggregation in the peripheral zone (Fig.4-14) these histological changes confirmed with (Cecil *et al*., 2008) which showed defect and sever damage in gray mater.

4.1.1.4: The histological Results of Brain Treated Group with (Lead +*Nerium oleander*) after 30 days orally administration

The histological results of brain after treated with lead and *Nerium oleander* compound, noted acute blood hemorrhage exactly in peripheral zone of gray matter, the blood hemorrhage appeared as wide elongated space filled with blood, so, showed hemolysis spots in the brain tissue similar to tissue necrosis, so, the tissue sections showed high cellular proliferation of inflammatory cells in the infected parts of brain, the tissue section was didn't have normal distribution of glial cells in the brain tissue (Fig.4-15), the tissue section didn't showed normal shape of cell nuclei in both nerve and glial cells in the brain tissue. These result may be due to oxidative effects on brain, these finding was agreement with (Buraimoh *et al*., 2012) which noted the metal exposure caused necrosis (neuro-degeneration).

The histological results showed the tissue section of brain have long band from abnormal cells in the middle regions of gray matter of brain, these cells have dark spherical or oval nuclei acidophilic cytoplasm and showed prominent space between cells (Fig.4-16), so, showed blood congestion in different location of brain tissue, prominent space belong the gray matter of brain tissue and wide spots of blood, these results may be due to high toxicity of lead which caused blood congestion and damage of brain, these results confirmed with (Patnaik *et al*., 2011) which noted

nerve cell degeneration, vacuolization and congestion of blood in brain after exposure to lead.

The tissue section of brain noted abnormal cellular proliferation in the wide regions of brain tissue, in this regions noted the cells have prominent space around the nuclei, some cells didn't have prominent nuclei (Fig.4-16). This histological change may be because eof the free radicals interact with molecular contents of the nucleus caused degenerative in nerve cells and brain tissue, this result confirmed with (Patnaik *et al*., 2011) which showed the free radicals caused damage in cellular component on nucleus and brain tissue.

The histological results showed the tissue section of brain have rounded lesion of necrosis, so, showed low cellular proliferation of glial cells in the brain tissue (Fig.4-11), these histological results agreement with (Loganathan *et al*., 2006) which noted severe necrosis of neuronal cells in cerebrum because lead acts directly on the cerebral vasculatures including blood-brain barrier and caused cerebral edema.

The tissue section of brain noted prominent space filled with glial cells in the brain tissue, also in this region huge necrosis appeared (Fig.4-17), these histological results coincide with (Loganathan *et al*., 2006) which noted were seen infiltration For the inflammatory cells and necrosis or death of neural cells.

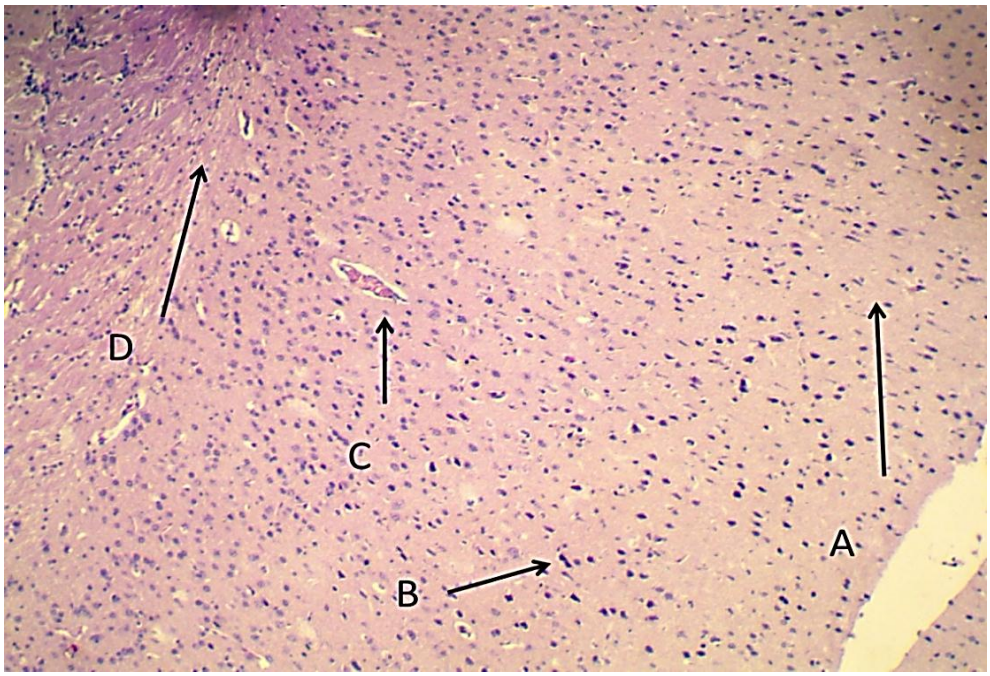


Fig (4-1): Transverse section of cerebral cortex in control group which showed A- Gray mater , B- Glial cell, C- Blood vessel , D- White mater . **H&E** stain

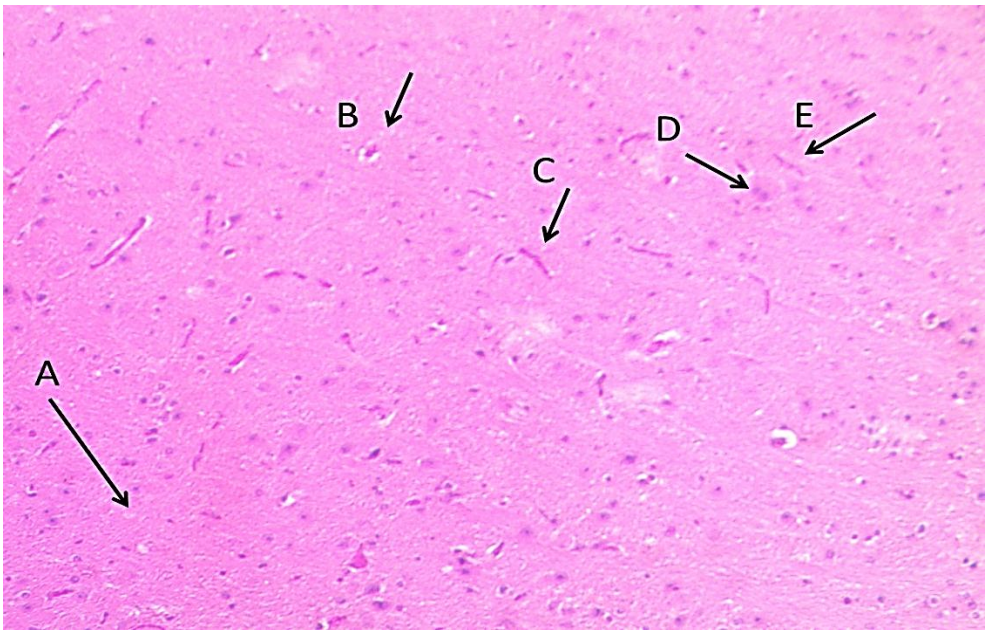


Fig (4-2): Transverse section of cerebral cortex in control group which showed the A- Gray mater, B- Nerve cell body, C- Nerve cell, D- Axon, E-Nerve cell . **H&E** stain 10X.

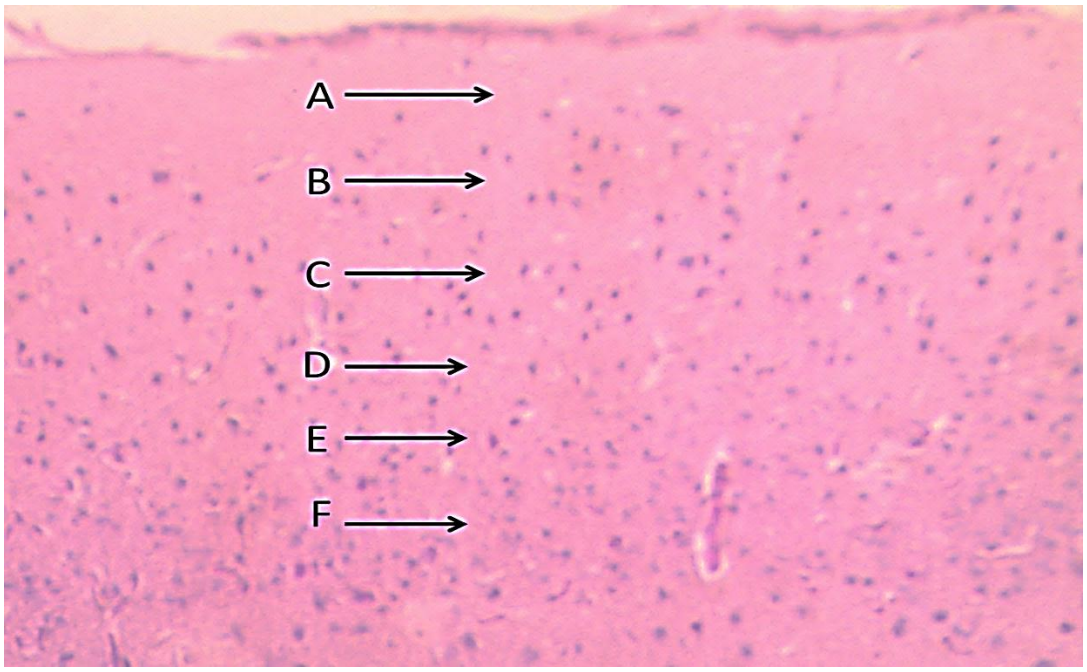


Fig (4-3): Transverse section of cerebral cortex in control group which showed the A- Molecular layer, B- External granular layer, C- Pyramidal layer, D- Inner granular layer, E- Ganglionic layer, F- Multiform layer . **H&E** stain 10X.

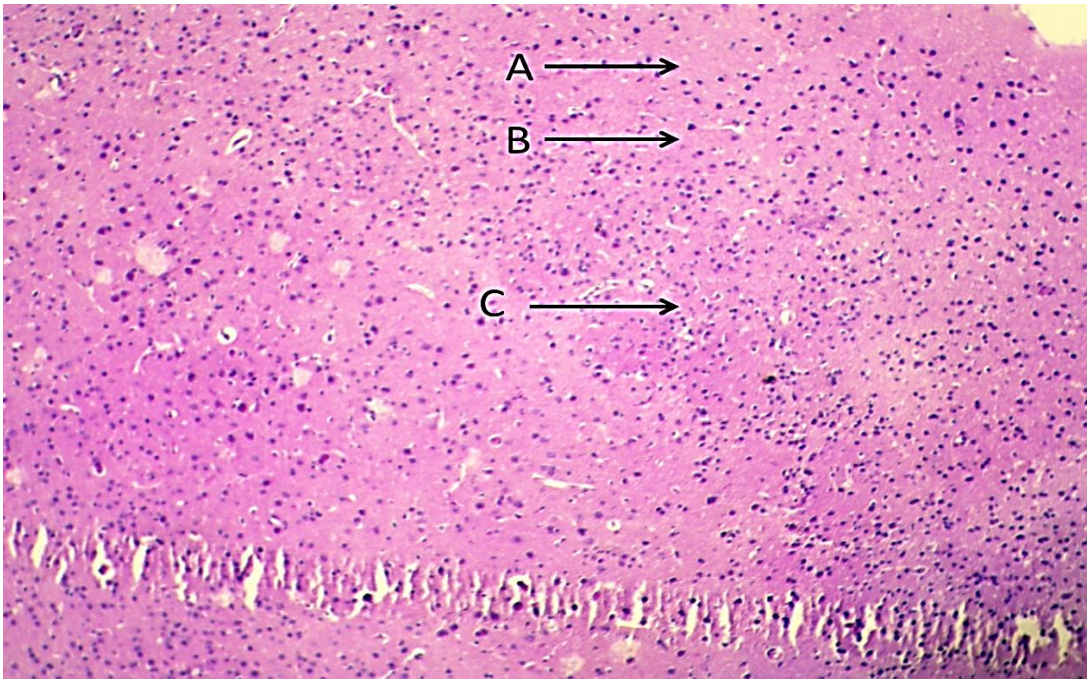


Fig (4-4): Transverse section of cerebral cortex in control group which showed the A- Molecular layer, B- Purkinje cells , C- Granular layer. **H&E** stain 10X.

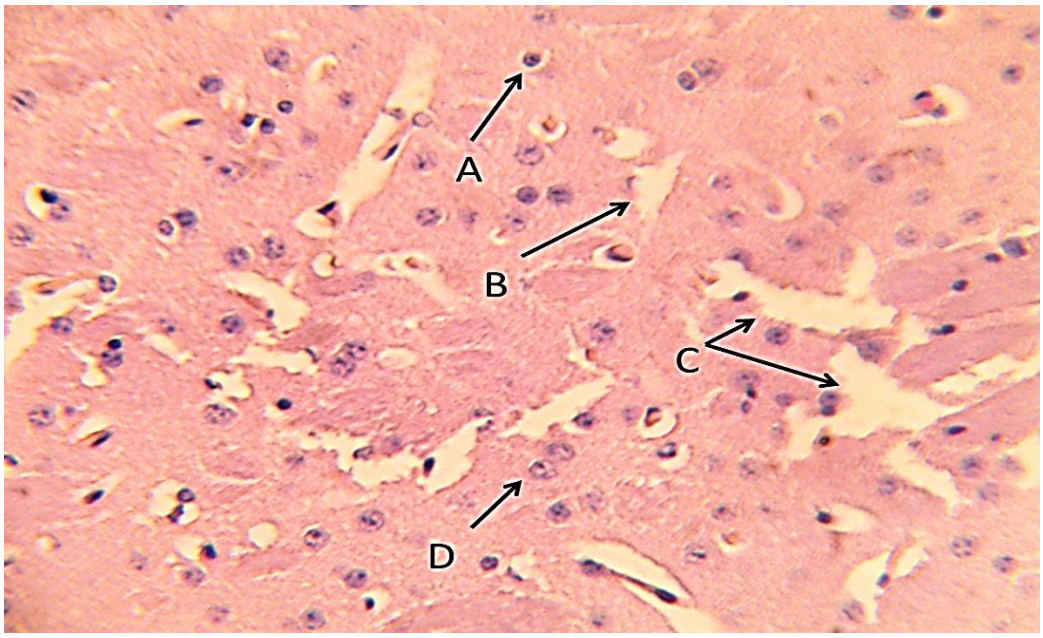


Fig (4-5): Transverse section of cerebral cortex in treated group with *Nerium oleander* which showed A-Cell have spherical nuclei , B- Necrosis, C-Tissue degeneration , D- Cell hyper trophy. **H&E** stain 40X.

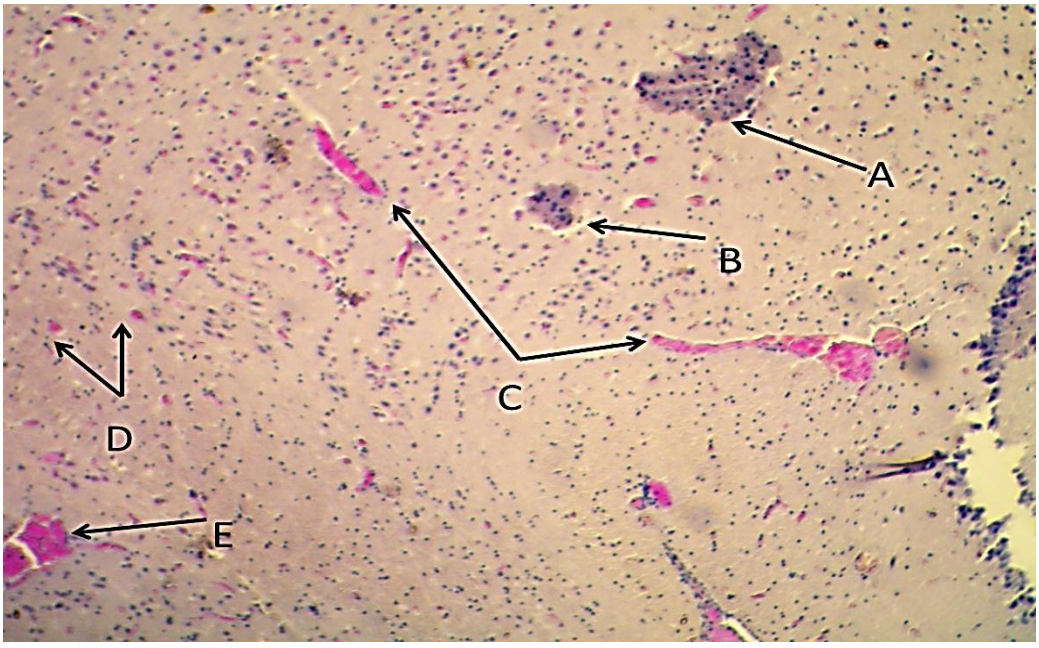


Fig (4-6): Transverse section of cerebral cortex in treated group with *Nerium oleander* which showed A-Cellular proliferation , B- Apoptosis, C-Blood vessel congestion, D- Blood congestion E- Cystic dilation filled with blood. **H&E** stain

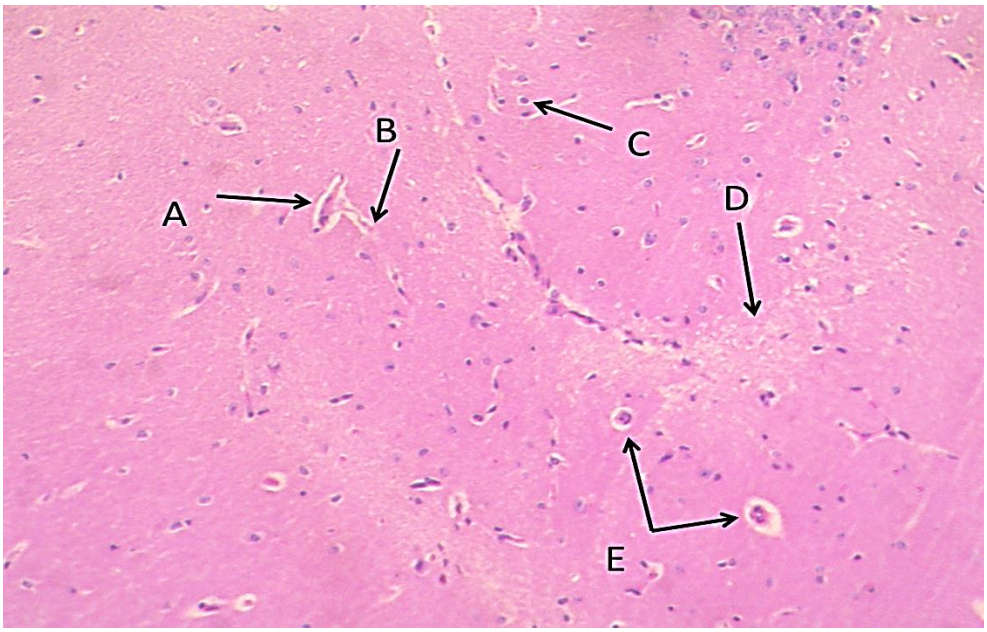


Fig (4-7): Transverse section of cerebral cortex in treated group with *Nerium oleander* which showed A- Nerve cell body, B- Dendrite , C- Glial cell have oval nuclei , D- Necrosis, E- Glial cell. **H&E** stain 10X.

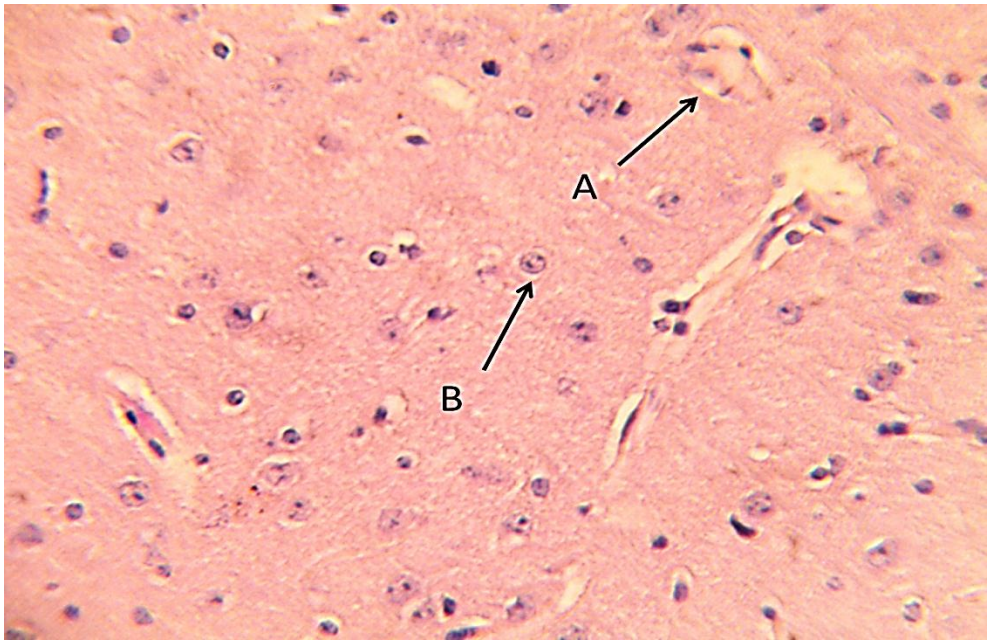


Fig (4-8): Transverse section of cerebral cortex in treated group with *Nerium oleander* which showed A- Spot of Apoptosis, B- Glial have dark nuclei . **H&E** stain 40X.

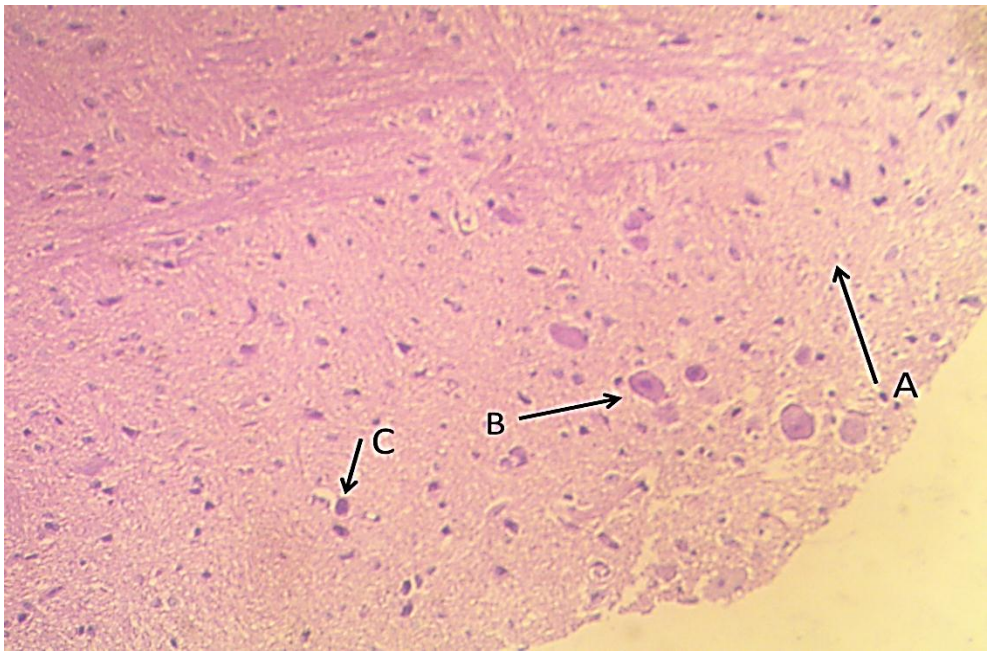


Fig (4-9): Transverse section of cerebral cortex in treated group with *Nerium oleander* which showed A- Empty space in tissue, B- Nerve cell, C- Glial cell. **H&E** stain 20X.

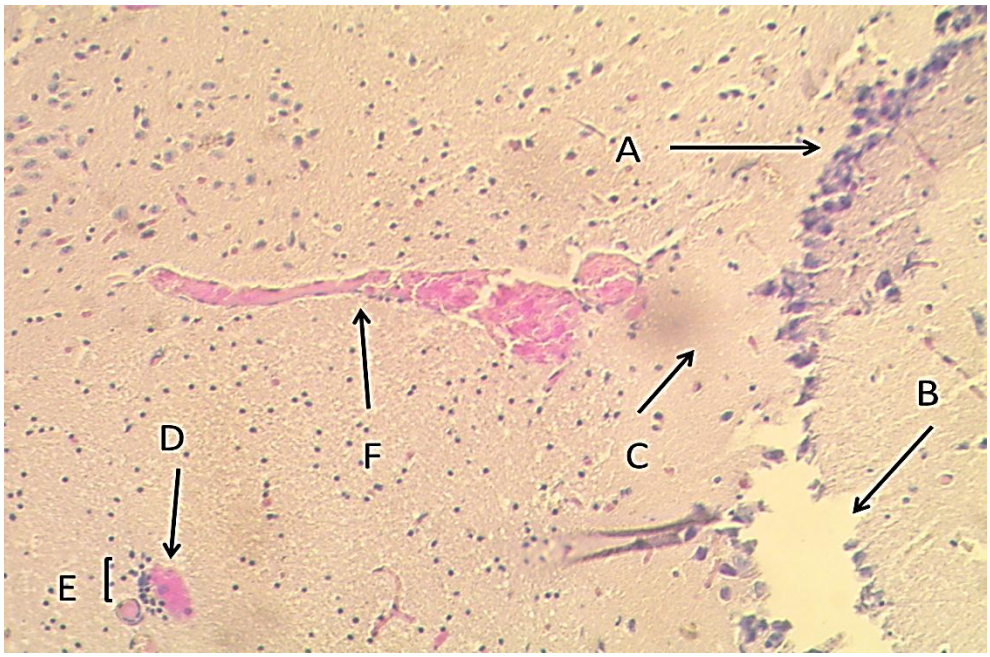


Fig (4-10): Transverse section of cerebral cortex in treated group with *Nerium oleander* which showed A- Glial cellular proliferation, B- Necrosis, C- Destruction or acute degeneration in the peripheral regions, D- Blood congestion, E- Proliferation of inflammation, F- Blood vessel. **H&E** stain 20X.

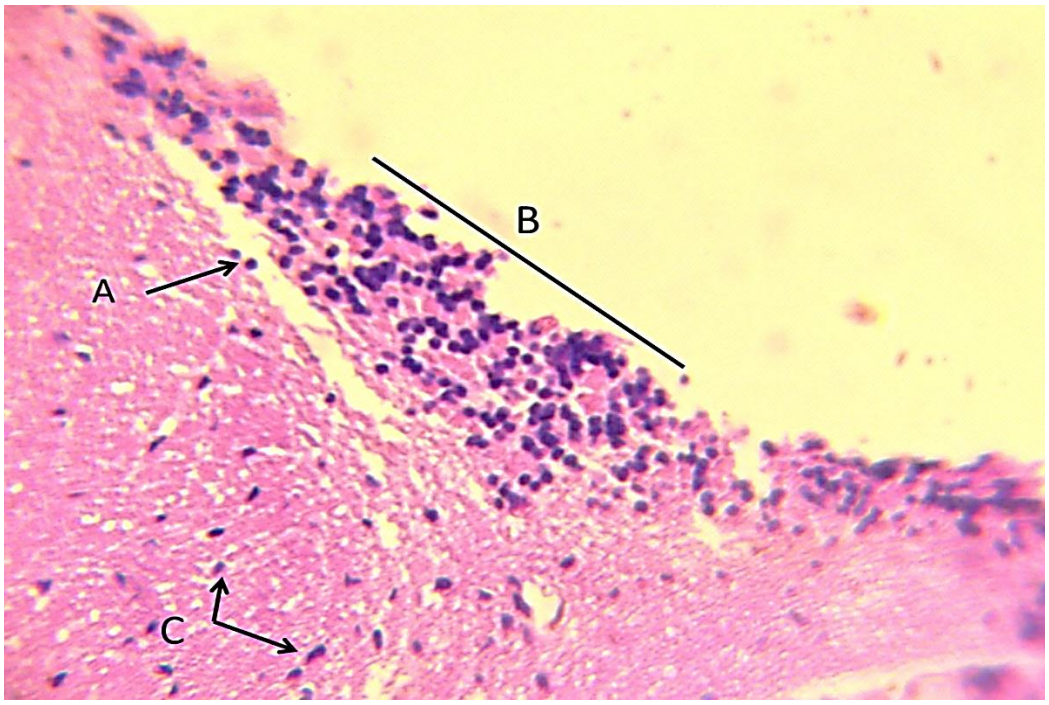


Fig (4-11): Transverse section of cerebral cortex in treated group with lead which showed A-Tissue degeneration, B- Cellular proliferation, C- Glial cell. **H&E** stain 20X.

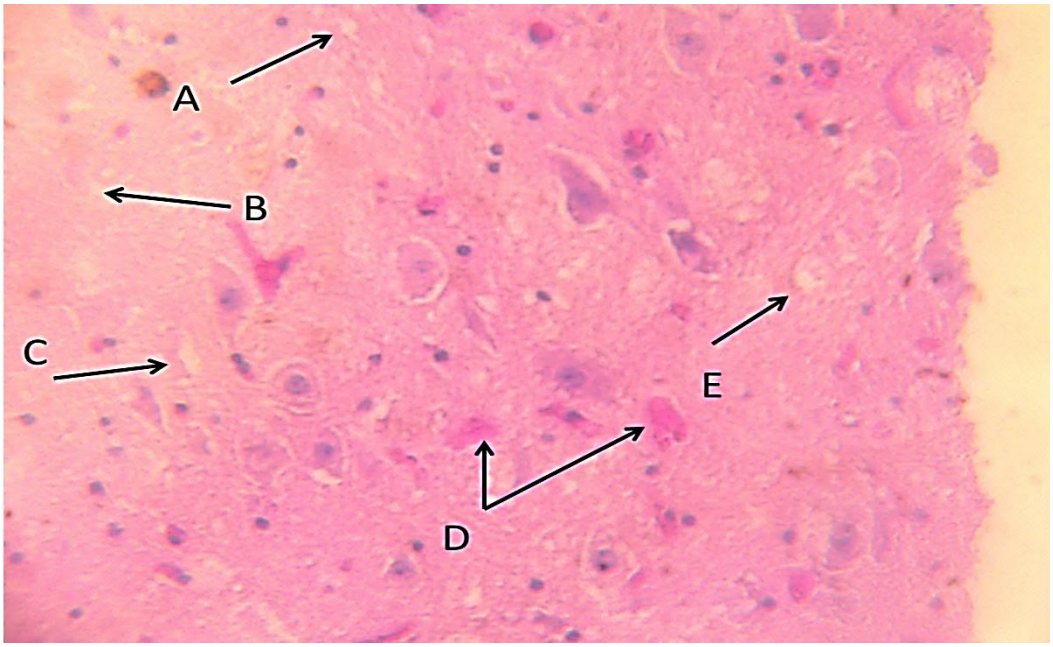


Fig (4-12): Transverse section of cerebral cortex in treated group with lead which showed A-Necrosis, B- Acute degeneration, C- Necrosis, D-Space filled with blood, E- Vacuole. **H&E** stain 40X.

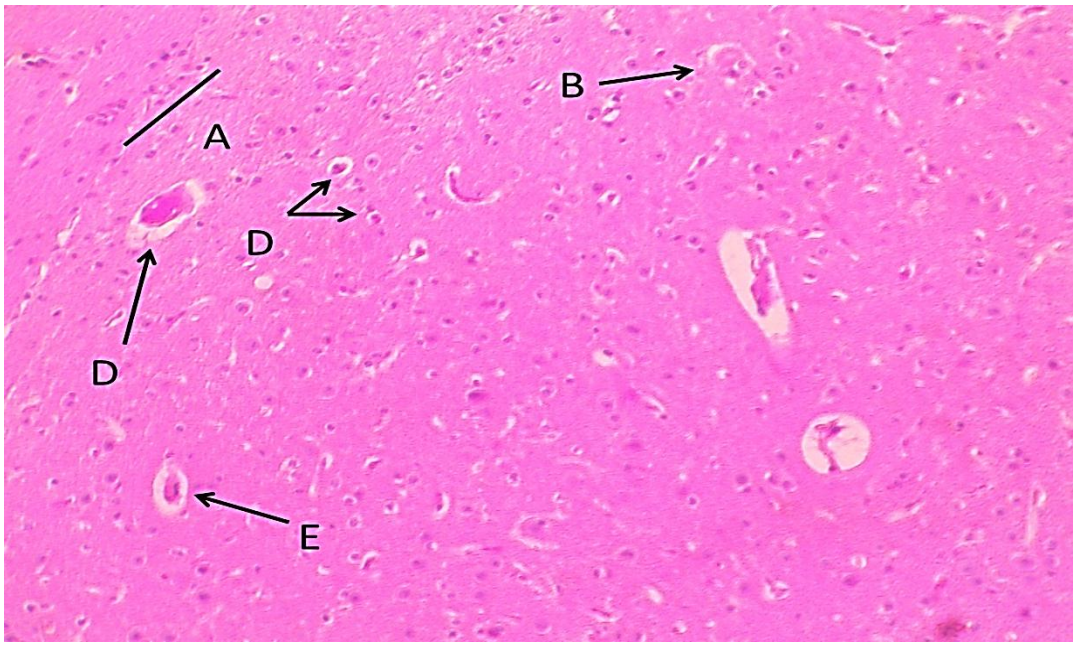


Fig (4-13): Transverse section of cerebral cortex in treated group with lead which showed A-Cellular proliferation, B- Blood congestion, C-Blood vessel congestion, D-Glial cell, E-Blood hemorrhage . **H&E** stain 20X.

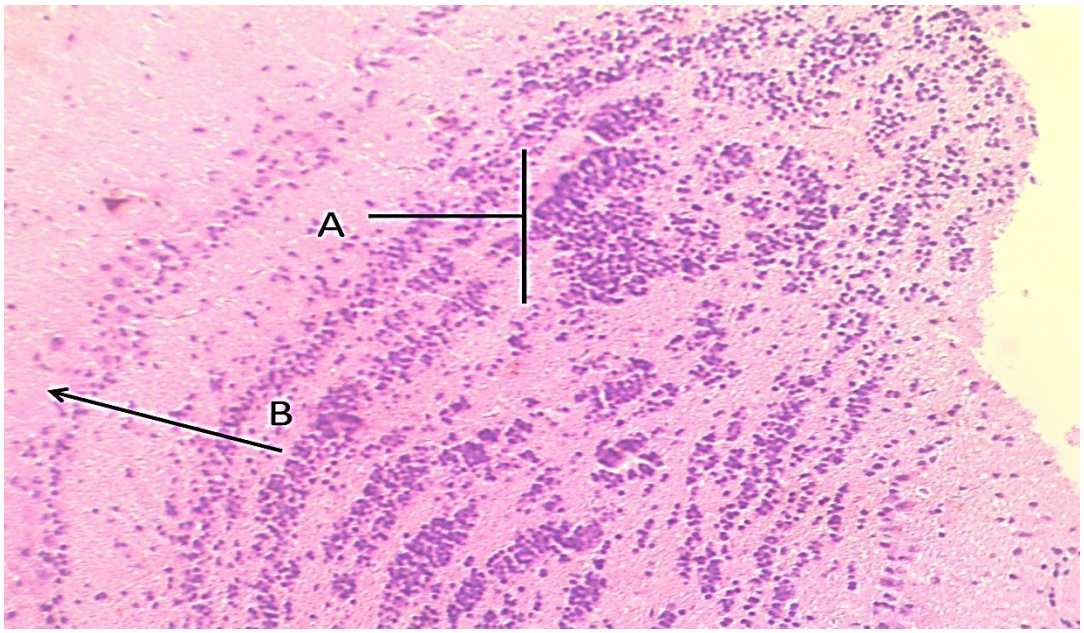


Fig (4-14): Transverse section of cerebral cortex in treated group with lead which showed A-Abnormal cellular proliferation, B- Abnormal layers of gray mater. **H&E**

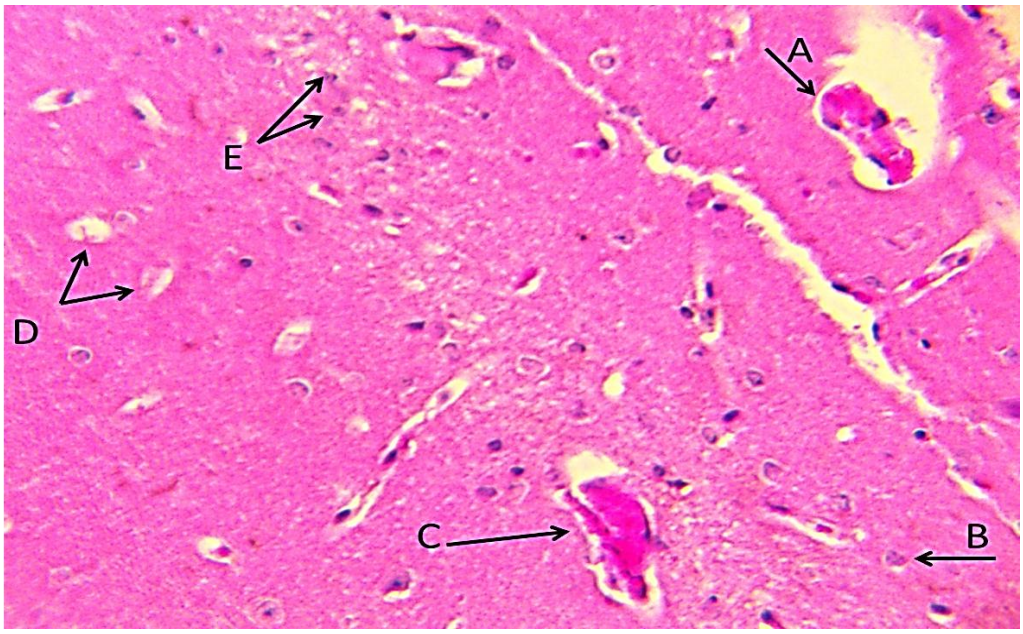


Fig (4-15): Transverse section of cerebral cortex in treated group with compound which showed A- Blood hemorrhage, B- Glial cell , C- Blood hemorrhage appeared as wide elongated space, D- Necrosis, E- Inflammatory cell. **H&E** stain 40X.

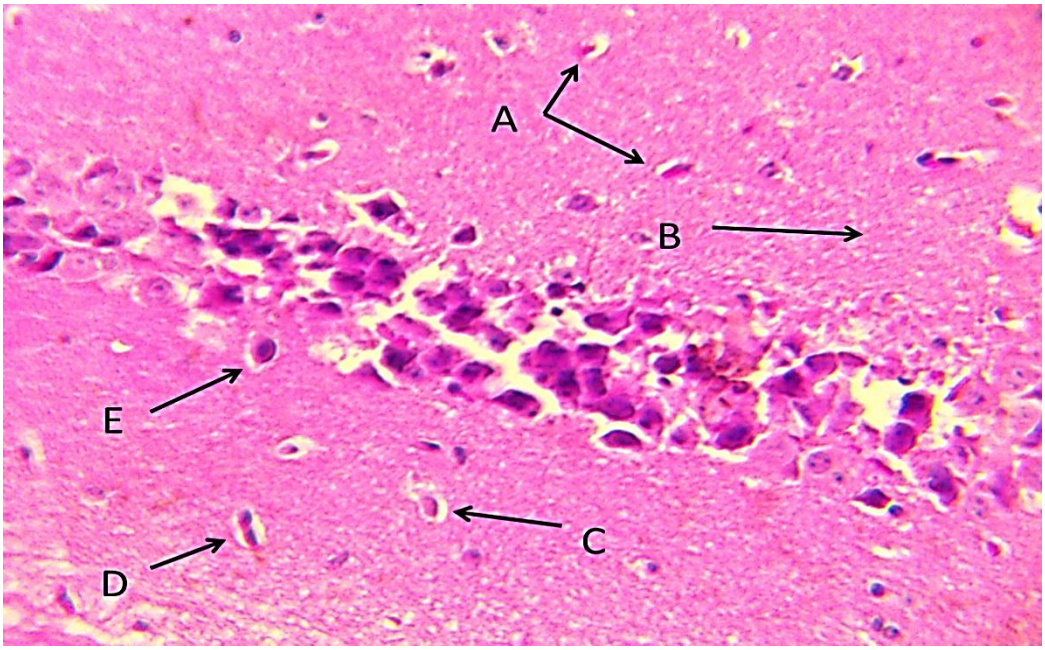


Fig (4-16): Transverse section of cerebral cortex in treated group with compound which showed A-Blood congestion, B- Prominent in tissue section, C- Nerve cell body, D- Nerve cell have multi nuclei, E- Nerve cell have dark nuclei. **H&E** stain

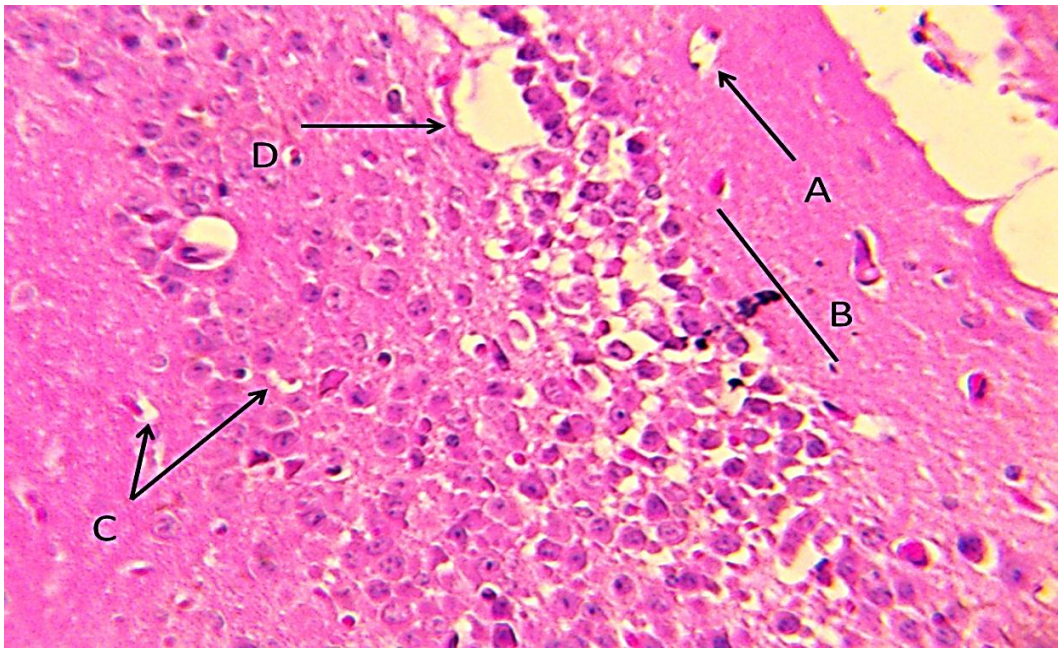


Fig (4-17): Transverse section of cerebral cortex in treated group with compound which showed A-Necrosis, B- Macrophage plasma cell, C-Small space, D-Huge necrosis. **H&E** stain 40X.

4.1.2: The Histological Results of Small intestinal

4.1.2.1: The Histological Results of Control Group

The current study showed the histological structures of small intestine in control group noted the wall of small intestine composed of four layers which included the first internal layer called the mucosa, the epithelial layer was simple columnar epithelial have (5.80±0.178µm),(Table 4-1) in thickness that rest on the basal lamina. The second layer called submucosal layer contain blood vessels and connective tissue fibers, the third layer called muscularis external was a region of muscle adjacent to the submucosa and outer layer consist from loose connective called serosa (outermost layer) of the intestine (Fig.4-18).

Intestinal villi (singular: villus) were tiny finger-like projections that protrude from the mucosa. Each villus have many microvilli (singular: microvillus), each of which were much smaller than a single villus. Villi responsible of increased the surface area of the intestine canal. This increased surface area be available for absorption. The villi are connected to blood vessels that carry the nutrients away in the circulating blood (Fig. 4-19).

4.1.2.2: The histological Results of Small intestinal Treated Group with *Nerium oleander* after 30 days orally administration

The histological result of the small intestine after treated with *Nerium oleander* showed the thickness of the epithelial layer was (4.71±0.120µm) which have significantly decreased compared with the control group as shown in (Table 4-1).

The tissue section of intestine noted the mucosal layer have prominent histological changes after the treated time, the tissue changes included exfoliated the epithelial layer that lined the internal surface of intestinal lumen and most epithelial cells aggregation as wide spots between the villi (Fig.4-20).

The other tissue section in terminal portion of small intestine showed epithelial cells hypertrophy and the epithelial cells have large vacuoles in their cytoplasm, the outer surface of villi appeared smoothly(Fig.4-21). These histological changes of small intestine may be due to the toxic agent lead to direct effects on smooth muscles of intestine, these results were confirmed with (Jubb *et al*., 1995) which noted the *Nerium oleander* toxicity which caused hypertrophy of which cell in intestine, destruction in villi and degeneration observed in the organs.

The tissue section noted abnormal structures of submucosal layer under the mucosal layer, prominent space belong the internal core of villi, most tissue section showed separated the base of villi from the under muscular layer in the wall of small intestine, so, showed blood congestions under the basal portions of villi (Fig.4-22), these histological changes agreement with (Akhtar *et al*., 2014) which noted the *Nerium oleander* affect on intestine caused congestion and hemorrhage.

The tunica muscularis was separated into three thin layers, the tissue section appeared prominent aggregations of inflammatory cells exactly between the tissue layers of small intestine (Fig.4-23). These histological result of small intestine after treated with *Nerium oleander* were coincide with (Jubb *et al*., 1995) which showed the *Nerium oleander* affect on small intestine caused chronic active inflammation in the muscularis.

The histological results showed the epithelial layer that covering the outer surface of villi have dark oval nuclei was elevated from the basal portion of epithelial cells, most tissue section showed prominent distribution belong the epithelial layer of small intestine (Fig.4-21). The submucosal layer appeared spongy structures have irregular empty spaces, the muscularis was very thin with prominent oval nuclei.

4.1.2.3: The Histological Results of Small intestinal Treated Group with Lead after 30 days orally administration

The histological result of small intestine after treated with lead showed the thickness of the epithelial layer was (6.21±0.163µm),(Table4-1) which didn't have significant increased compared with the control group.

The histological section of small intestine in mice after treated with lead for thirty days of experimental time note prominent histological changes in the tissue structures in the wall of small intestine.

The histological result noted all tissue layer of small intestine have prominent changes which included the mucosal layer of small intestine have prominent destruction in epithelial layer. The epithelial layer that covering the outer surface of villi in small intestine have prominent vacuoles, and most epithelial cells were without nuclei, the epithelial layer of some villi was completely isolated from the underlying tissue layers (Fig.4-24). These results may be due to the accumulation of lead in tissue caused dysfunctional in intestine wall this result confirmed with (Yuan

et al., 2014) which noted the lead caused intestinal epithelium injury and epithelial cells.

The core of villi have prominent degeneration, the sub mucosal layer was very thin and disappeared from most villi, the basal portions of villi were completely separated in the underlying connective tissue. The tissue section showed inflammatory cells aggregation between at a portion of villi (Fig.4-25). These histological changes may be due to the lead poisoning lead to this injury, these results agreement with (Ahmed *et al*., 2014) which showed strong inflammatory reactions in intestine after exposure of lead.

The muscular layers were isolated from the inner layers of intestinal wall, the muscular layer have abnormal cellular proliferation, the tissue section of small intestine showed prominent blood hemorrhages between sub mucosa and muscular layers of intestinal wall, the outer layer was very thin and adhesion with muscular layer (Fig.4-26), the tissue pictures showed the internal lumen of small intestine was filled with exfoliated epithelial cells. These histological result may be due to the exposure of lead caused hemorrhage, damaged in epithelial cells, cellular propagation were result coincide with (Ahmed *et al*., 2014) which noted hemorrhage, defect in muscular layer and epithelial cells.

The histological results of small intestine showed high cellular infiltration in the basal portion of villi, the smooth mucosal cells in the intestinal wall have prominent elongated nuclei, the most tissue section of small intestine showed abnormal aggregation of inflammatory cells between the basal sections of villi (Fig.4-26). These histological changes in intestine after treated with lead agreement with (Yuan *et al*., 2014) which showed proliferation inflammatory cells, prominent in epithelial cells and injury in villi.

4.1.2.4: The histological Results of Small intestine Treated Group with (Lead+*Nerium oleander*) after 30 days orally administration

The histological result of small intestine after treated with *Nerium oleander* and lead showed the thickness of epithelial layer have significant decreased (5.21±0.151µm),(Table 4-1) in compared with control group.

The histological results of intestine after treated with compound lead and *Nerium oleander* solution for thirty days, which showed prominent histological changes in the wall of small intestine, the tissue section showed separated the tissue layers that

composed of the small intestine wall, completely separated submucosal layer from tunica muscularis, so, noted wide space between submucosa and muscularis layers (Fig.4-27). These histological changes agreement with (Adiguzel and Kalender, 2015) which showed the lead exposure to the small intestine caused separated the layers of the intestine and degeneration in layers and epithelium.

Most tissue section of small intestine in the treated mice have prominent aggregation of inflammatory cells at the base portion of villi, no clear the core of villi, the villi covered by columnar or high cuboidal epithelia which differ from the tissue section of intestine wall in control group (Fig.4-28). These histological results may be due villi uptake of the of lead which caused disorder in villi and proliferation of inflammatory cells the result agreement with (Abdel-Warith *et al*., 2020) which showed inflammatory cell infiltration and injury in villi.

The epithelial cells have prominent spherical dark nuclei, the tissue section showed hypertrophy in the most of villi, which lead to modification in the shape of epithelial cells (Fig.4-29), all tissue section didn't have microvilli on the outer surface of villi, most tissue section didn't have normal villi, the residual villi didn't have normal epithelial layer. These results of intestine after treated with lead may be due to the sensitive of intestine epithelia that lead to toxicity which caused damage in villi, this result confirmed with (Abdel-Warith *et al*., 2020) which showed alteration in epithelial cells and villi.

The epithelial cells exfoliated and accumulation between the abnormal villi, the base of affected villi completely separated from underling connective tissue, the result showed prominent vacuoles in the cytoplasm of the epithelial cells, prominent degeneration in the wall of small intestine and high inflammatory cells proliferation (Fig.4-29). These results referred to acute toxicity of lead to degeneration in connective tissue, this result coincide with (Adiguzel and Kalender, 2015) which observed that exposure to lead caused damage and dysfunction in the layer of small intestine and villi.

The results showed prominent necrosis lesions seen in the effected villi, so high blood congestion in some sections of small intestine, but the other have prominent blood hemorrhage. Most tissue section showed completely destruction of the apical portion of villi and remained the base portion of villi which weakly adhesion with tissue layers (Fig.4-27). These results may be due to accumulation of free radicals of lead caused damage of blood vessels and villi, these results confirmed with (Olaleye

et al., 2007) which noted the free radicals of lead caused destruction in villi and blood congestion.

Table (4-1) : Measurement of epithelial layers of small intestine in study group in mice M± S.D

Treatment	Control	Lead	*Nerium oleander*	Compound
Thickness layer	5.80±0.178[a]	6.21±0.163[a]	4.71±0.120[c]	5.21±0.151[b]

Different letters vertically indicate the existence of significant differences between the averages at the possibility of (0.01).

(Table4.1-1) Results of ANOVA analysis of small intestine of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
Epithelial layer	Between Groups	65.074	3	21.691	18.046	.000
	Within groups	235.585	196	1.202		
	Total	300.659	199			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Nerium oleander	50	4.7180		
Compound	50		5.2104	
Control	50			5.8048
Lead	50			6.2168
Sig.		1.000	1.000	.062

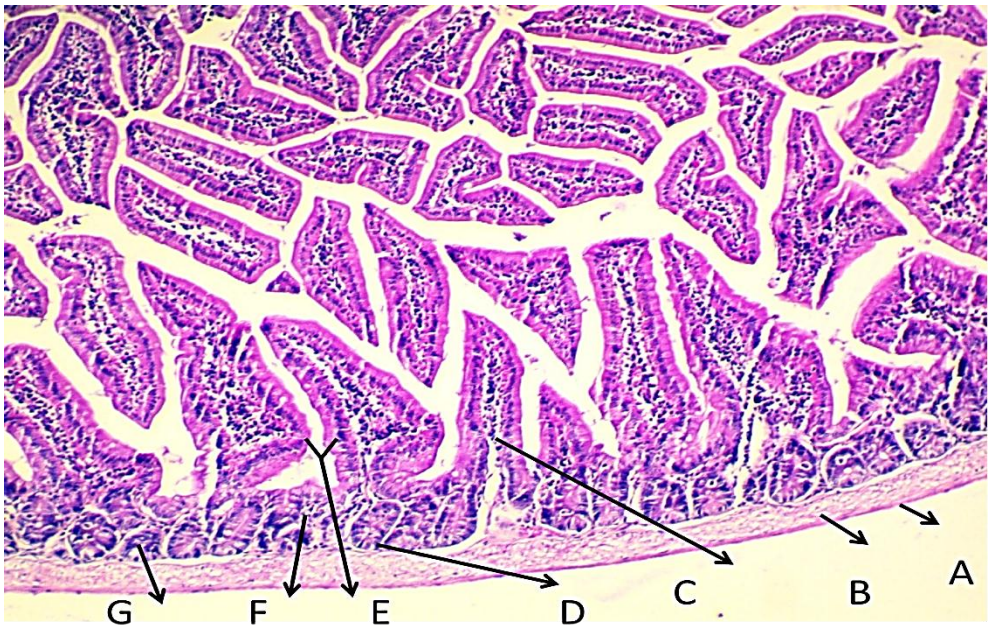


Fig (4-18): Transverse section of small intestine in control group which showed A-Serosa , B-Muscularis, C-Simple columnar, D-Sub mucosa, E-Villi, F-Blood vessel, G-Intestinal gland. **H&E** stain 20X.

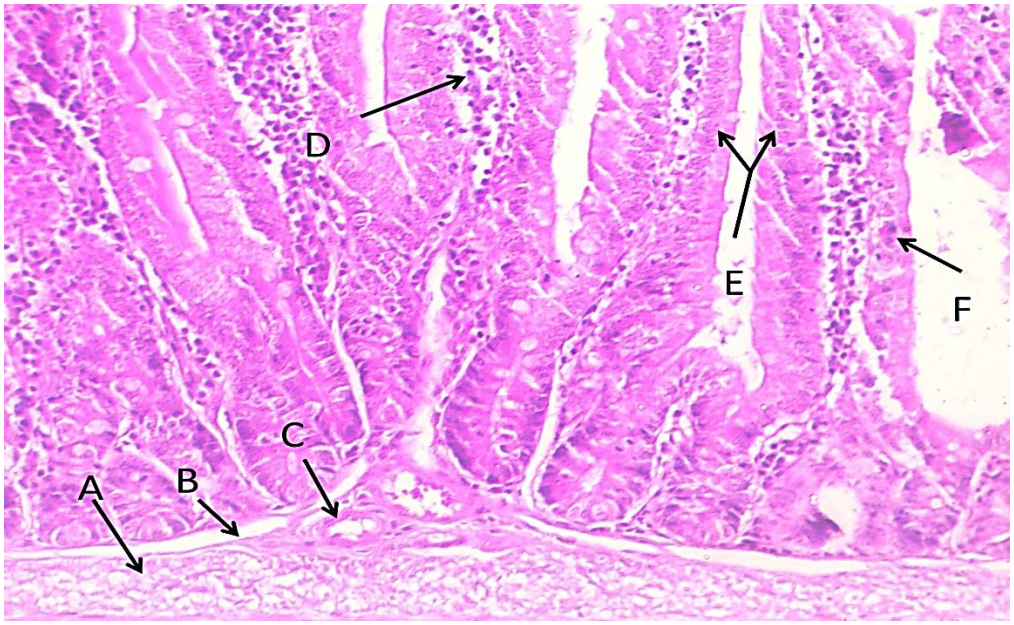


Fig (4-19): Transverse section of small intestine treated group with control group which showed A-Muscular layer, B- Sub mucosa, C-Blood vessel, D-Core of layer, E-Villi, F-Epithelia. **H&E** stain 40X.

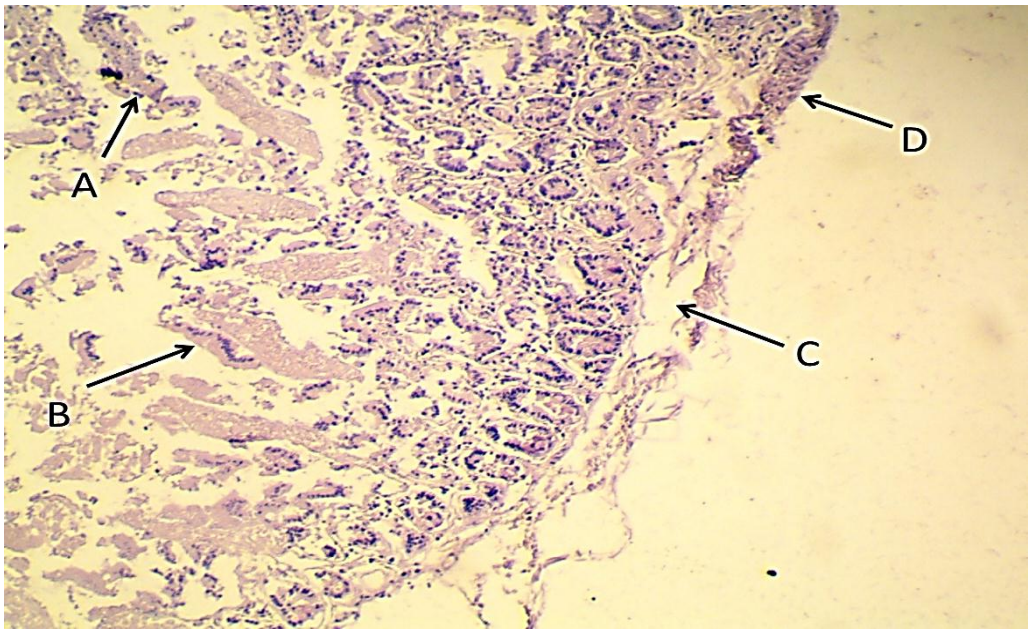


Fig (4-20): Transverse section of small intestine in treated group with *Nerium oleander* which showed A-Destruction villi, B- Exfoliated epithelia, C-Prominent damage in intestinal wall, D-Serosa. **H&E** stain 20X.

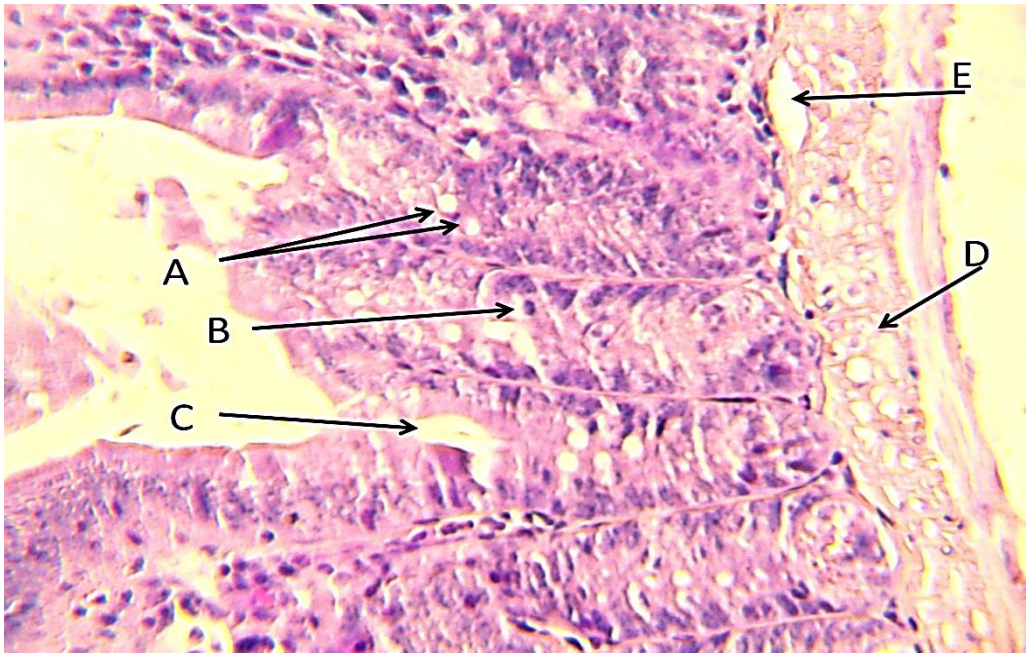


Fig (4-21): Transverse section of small intestine in treated group with *Nerium oleander* which showed A-Empty vacuole, B- Plasma cell lymphocytosis, C-Epithelia destruction, D-Weakness of muscle layer, E- Wide space. **H&E** stain

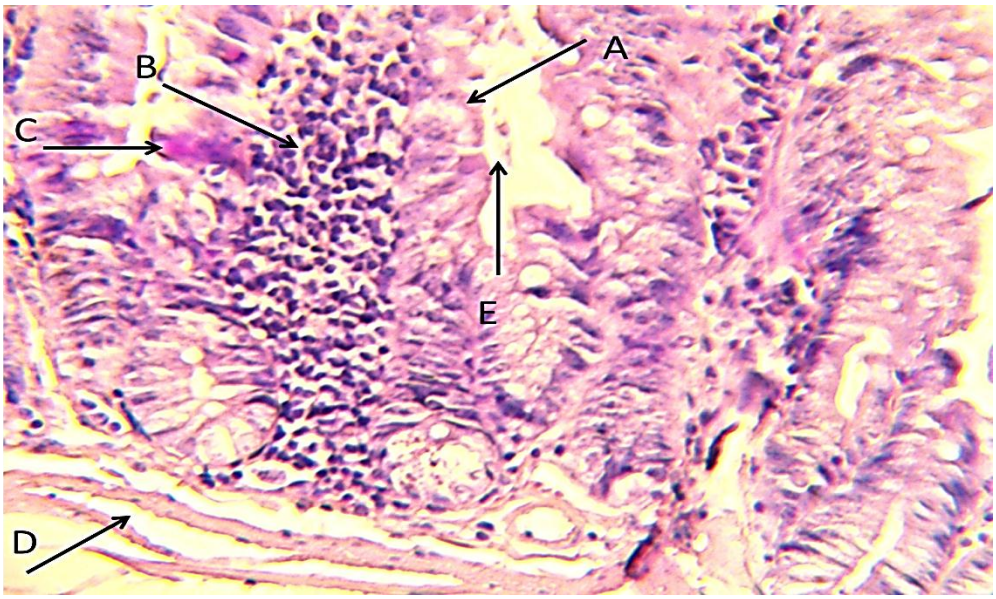


Fig (4-22): Transverse section of small intestine in treated group with *Nerium oleander* which showed A-Epithelial destruction, B- Lymphocytosis plasma cell infiltration, C- Blood congestion, D- Weakness muscular layer, E- Prominent space. **H&E** stain 40X**.**

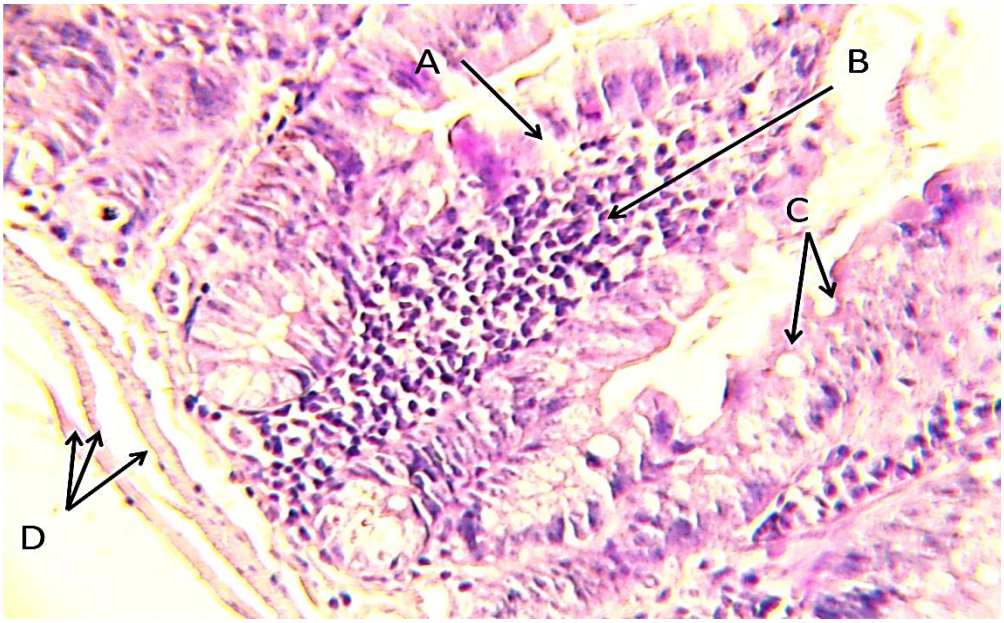


Fig (4-23): Transverse section of small intestine in treated group with *Nerium oleander* which showed A- Prominent destruction, B- Cellular infiltration, C- Vacuole, D- Completely separated three muscular layer . **H&E** stain 40X.

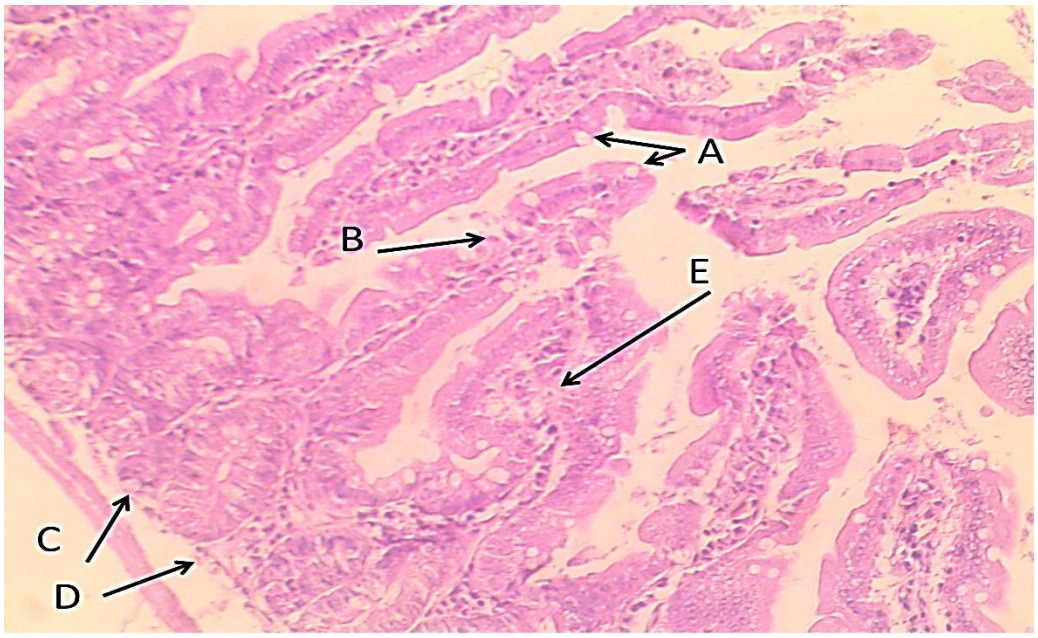


Fig (4-24): Transverse section of small intestine in treated group with lead which showed A- Goblet cell, B- Epithelial destruction, C- Isolated muscular layer, D- Separated of villi, E- Epithelial cells without nuclei. **H&E** stain 20X.

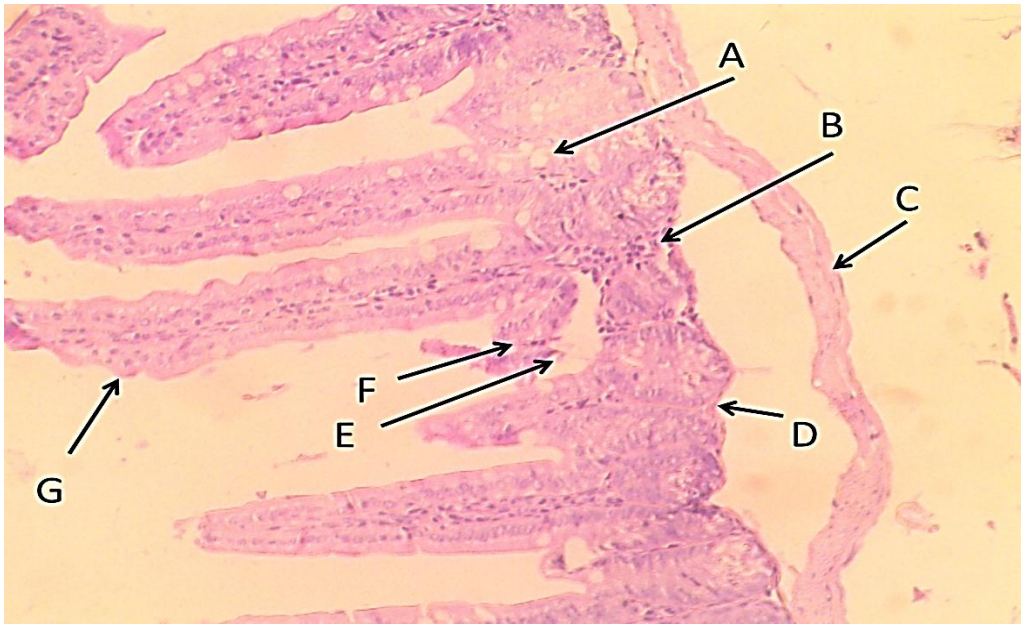


Fig (4-25): Transverse section of small intestine in treated group with lead which showed A- Goblet cell, B- Inflammatory cells, C- Completely separated of muscular layer, D- Submucosa, E- Prominent degeneration, F- No core of villi, G- Abnormal epithelial. **H&E** stain 40X.

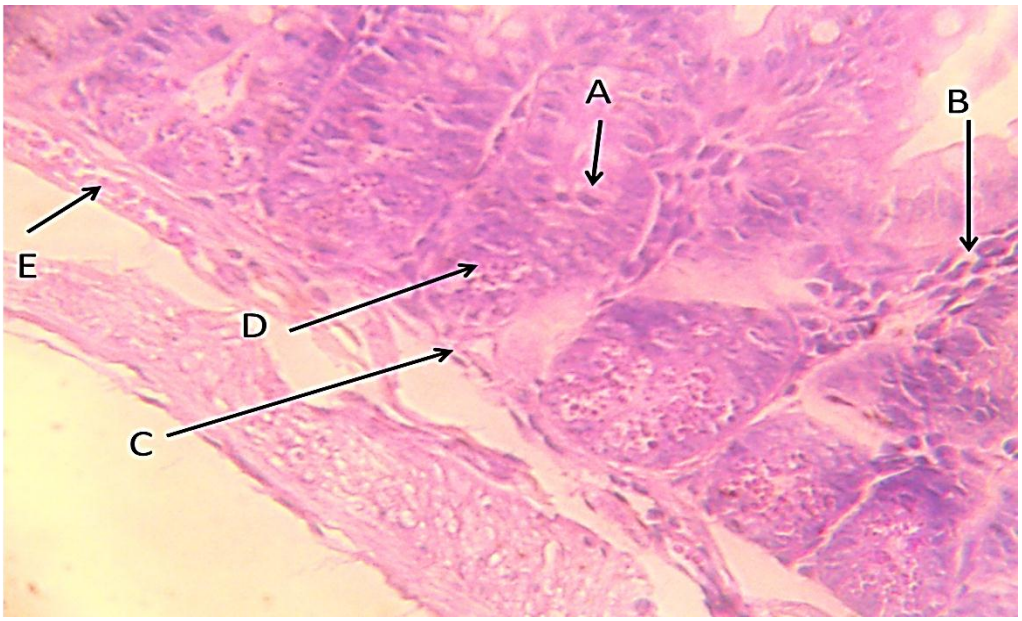


Fig (4-26): Transverse section of small intestine in treated group with lead which showed A- Columnar have elongated nuclei, B- Cellular proliferation, C- Villi lose their connection to each other, D- Exfoliated epithelial cells, E- Blood hemorrhage . **H&E** stain 40X.

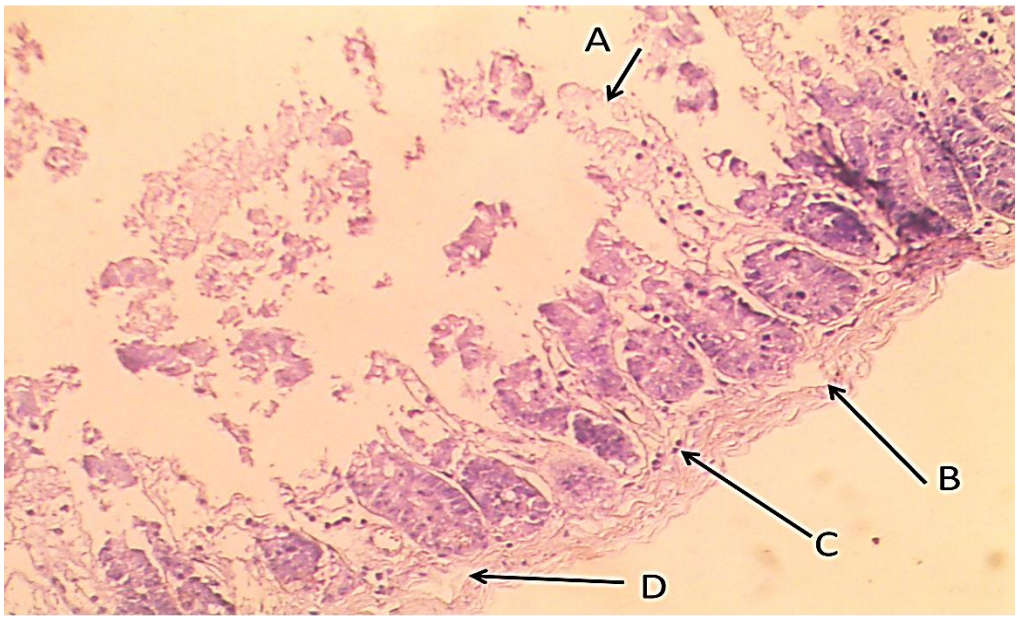


Fig (4-27): Transverse section of small intestine in treated group with compound which showed A- Necrosis, B- Separated tissue layer composed the small intestine, C- Dispersed or weak muscular layer, E- space between submucosa and muscularis layers. **H&E** stain 20X

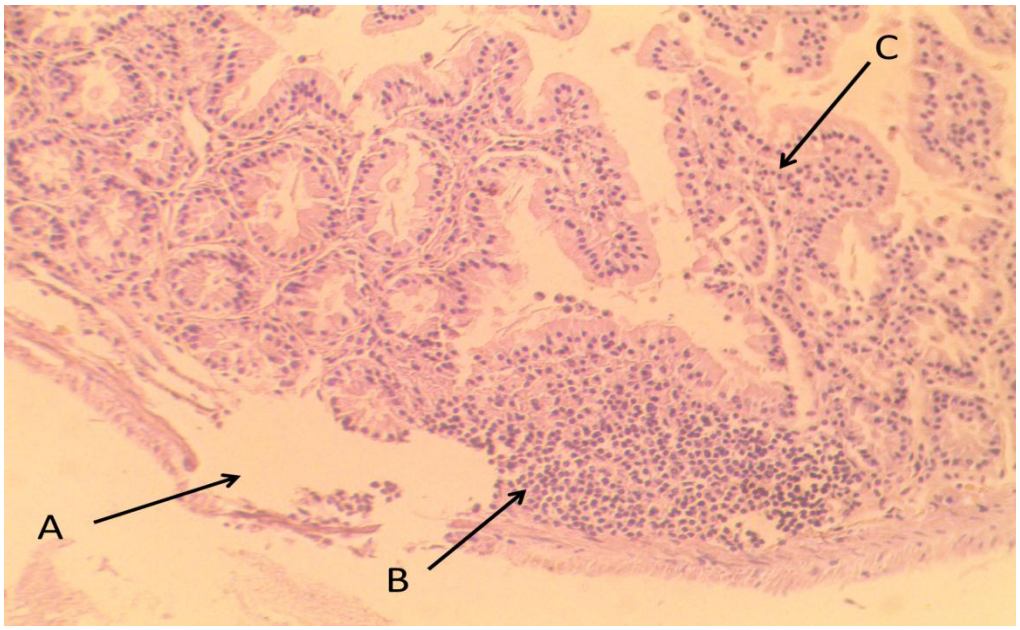


Fig (4-28): Transverse section of small intestine in treated group with compound which showed A- Prominent degeneration, B- Cellular infiltration, C- No core of villi. **H&E** stain 20X.

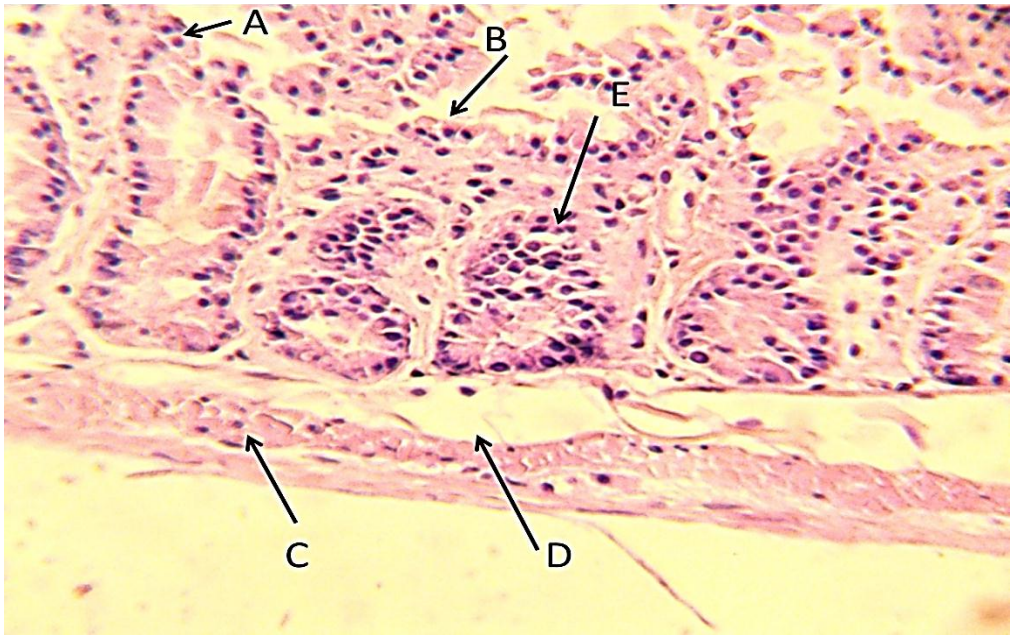


Fig (4-29): Transverse section of small intestine in treated group with compound which showed A-Epithelial destruction, B- Hypertrophy of villi, C- Muscular layer, D- Completely separated. **H&E** stain 40X.

4.1.3.: The Histological Results of Liver

4.1.3.1: The Histological Results of Liver of Control Group

The histological result of liver in control group showed the diameter of hepatocyte nuclei was (9.18± 0.208µm), the liver parenchyma composed of hepatic cords that consist from hepatocytes that arranged as long cordal structures, between hepatic cords showed the prominent sinusoids. The kupffer cells were normal distribution through the hepatic parenchyma. The liver was surrounding by thin connective tissue capsule Fig (4-30).

The current results showed the hepatocyte have spherical pigmented dark nuclei with acidophilic cytoplasm. The present result of liver tissue in the control group noted normal distributed of kupffer cells between hepatic cords. The histological result of liver in the control group appear normal blood sinusoid that line by normal endothelial layer (Figure 4-31).

4.1.3.2: The histological Results of Liver Treated Group with *Nerium oleander* after 30 days orally administration

Most tissue sections of liver after treated with *Nerium oleander* for thirty days of experimental time showed prominent tissue changes in the liver parenchyma, these changes included acute cellular destruction, acute blood hemorrhage, and in other regions showed blood congestions, the histological results of liver after treated time noted wide aggregation of inflammatory cells located arrounded the region of hemorrhage (Fig.4-32), the tissue sections showed irregular spaces may be due to acute hepatocyte destruction, these space filled with blood. These results may be due to the liver consider as first organ sensitive to the poisons of *Nerium oleander*. This result was similar to (Majeed, 2012) which noted that the liver after exposure to *Nerium oleander* which lead to degenerative in liver tissue, blood congestion and inflammatory proliferation.

Generally the liver parenchyma didn't have normal long hepatic cords, so, no founded the normal portal area, many necrosis lesions that seen in liver, most hepatocyte didn't have normal nuclei, the nuclei have pigmented or granular chromatic material was randomly distributed in the nuclei of hepatocyte (Fig. 4-33). This histological change agreement with (Farkhondeh *et al*., 2020) which showed in the tissue of liver after treated with *Nerium oleander* scattered necrosis, injury in hepatocyte and degenerative in portal area.

The histological result of liver after treated with *Nerium oleander* have significant decreased in diameter of hepatocyte nuclei (7.82±0.181µm),(Table 4-2) in compared with control group.

The tissue section of liver showed inflammatory cells among the hepatocyte, most hepatocyte have clear cytoplasmic vacuoles (Fig. 4-33). This result confirmed with (Farkhondeh *et al*., 2020) which explained the cross section of liver showed infiltration of inflammatory cells and damage in cytoplasmic hepatocyte.

The results showed the tissue section of liver have wide circular space contained RBCs and surrounded by inflammatory cells, the tissue section showed wide tissue destruction in the liver parenchyma, congestion of central vein (Fig.4-34). This result may be sever toxicity lead to these in liver was result coincide with (Omidi *et al*., 2011) which showed degeneration in tissue and blood congestion.

4.1.3.3: The Histological Results of Liver Treated Group with Lead after 30 days orally administration

The histological results of liver after exposure to lead showed no significantly increased in diameter of hepatocyte nuclei (9.92± 0.312µm),(Table 4-2) compared with control group.

The histological result of liver after treated with lead solution for thirty days of experimental time, showed the liver parenchyma have blood hemorrhages in the liver tissue with prominent degeneration, the tissue section showed clear space between hepatocyte of liver tissue, no, prominent hepatic cords, most hepatocyte didn't have normal nuclei, the tissue sections showed the hepatocytes have abnormal distribution in the liver parenchyma (Fig4-35). This result may be due to accumulation of the lead solution in the liver tissue caused disorder and injury in liver tissue. This result agreement with (Haouas *et al*., 2014) which showed degeneration in hepatocyte, injury in liver tissue and sever hemorrhage in liver.

The tissue section noted wide empty cystic dilation in the liver parenchyma, prominent necrosis lesions in different location of liver tissue, the histological result of liver noted abnormal portal area with prominent hypertrophy in many hepatocyte, most hepatocyte destruction may be because of the hepatocyte hypertrophy (Fig.4-36), so, the result noted necrosis lesions, the liver of treated mice with lead have prominent tissue degeneration which appeared as irregular spaces filled with blood,

aggregation of inflammatory cells nearly from the spaces, most hepatocyte were isolated from each other, the liver parenchyma didn't have long or normal hepatic cords. These histological results may be due to accumulation or increased amount of lead which caused infiltration of inflammatory cells, cellular necrosis and damage in portal area. This result agreement with (Andrade and Bhat, 2019) which showed the liver tissue after exposure to lead caused prominent necrosis and inflammation response.

The result noted the liver parenchyma have wide central vein filled with blood or congestion, the central vein surrounded by aggregation of inflammatory cells, the most hepatocyte destruction or swelling, the result appeared prominent tissue degeneration or necrosis (Fig.4-37,38). This result similar to (Haouas *et al*., 2014) which observed the liver tissue after exposure to lead poisoning caused the hepatocyte appear loss of their normal shape and central vein congestion filled with blood hemorrhage.

4.1.3.4: The Histological Results of Liver Treated Group with after (Lead+*Nerium oleander*) 30 days orally administration

The histological result of liver after administration to lead and *Nerium oleander* showed no significant increase in diameter of hepatocyte nuclei (9.09± 0.389µm),(Table 4-2) in compared with control group.

The histological section of liver after treated with compound solution of lead and *Nerium oleander* for thirty days of experimental time that showed liver parenchyma have acute degeneration as wide regions of hepatocyte destruction, the destruction regions appeared as irregular wide empty space and disappeared the long cordal structure (Fig.4-39). This result may be due to the toxicity of *Nerium oleander* which lead to these histological changes, these results were agreement with (Jabouri and Rasha, 2008) which showed degenerative in liver parenchyma and hepatocyte necrosis.

Abnormal portal area was irregular in shape filled with blood and surrounded by inflammatory cells proliferation, most hepatocyte have oval dark nuclei with acidophilic cytoplasm, some of cells loss their nuclei, the result showed many necrosis lesions in different location of liver parenchyma (Fig.4-40). This histological change may be because of exposure to pollution of lead which caused

decrease or increase in liver enzyme effects on the liver parenchyma, this result similar to (Ashrafizadeh *et al*., 2018) which showed obvious degenerative in liver tissue and inflammatory aggregation darkly nuclei of hepatocyte.

The tissue section showed acute blood hemorrhage as wide cystic dilation filled with blood, the peripheral zone of liver parenchyma was shattered, the hepatocyte loss their nuclei, prominent inter lobular ducts that showed belong the liver parenchyma (Fig.4-41). This result confirmed with (Ashrafizadeh *et al*., 2018) which noted the oxidative stress caused vesicular congestion with hemorrhage in liver tissue and damage in hepatocyte.

The histological results showed very long spaces near the abnormal portal area which filled with thick secretion and blood, the peripheral line have prominent empty vacuoles, this tubular space surrounded by high proliferation of inflammatory cells (Fig.4-42), so, showed wide necrosis lesions, the tissue section showed abnormal wide cystic dilation, have prominent Apoptosis spots. This result confirmed with (Taheri *et al*., 2013) which showed the liver tissue after exposure to *Nerium oleander* alteration in portal area and apoptotic cells lead to formation wide spots and tubular space.

Table (4.2) : Measurement of hepatocyte nuclei in study group in mice M± S.D

Treatment	Control	Lead	*Nerium oleandr*	Compound
Diameter	9.18 ± 0.208^{a}	9.92 ± 0.312^{a}	7.82 ± 0.181^{b}	9.09 ± 0.389^{a}

Different letters vertically indicate the existence of significant differences between the averages at the possibility of (0.01)

(Table4.2-1) Results of ANOVA analysis of hepatocyte nuclei of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
Hepatocyte nuclei	Between Groups	114.329	3	38.110	9.361	.000
	Within groups	797.931	196	4.071		
	Total	912.260	199			

Duncan

TRT		Subset for alpha = 0.01	
		1	2
Nerium oleander	50	7.8202	
Compound	50		9.0932
Control	50		9.1848
Lead	50		9.9230
Sig.		1.000	.052

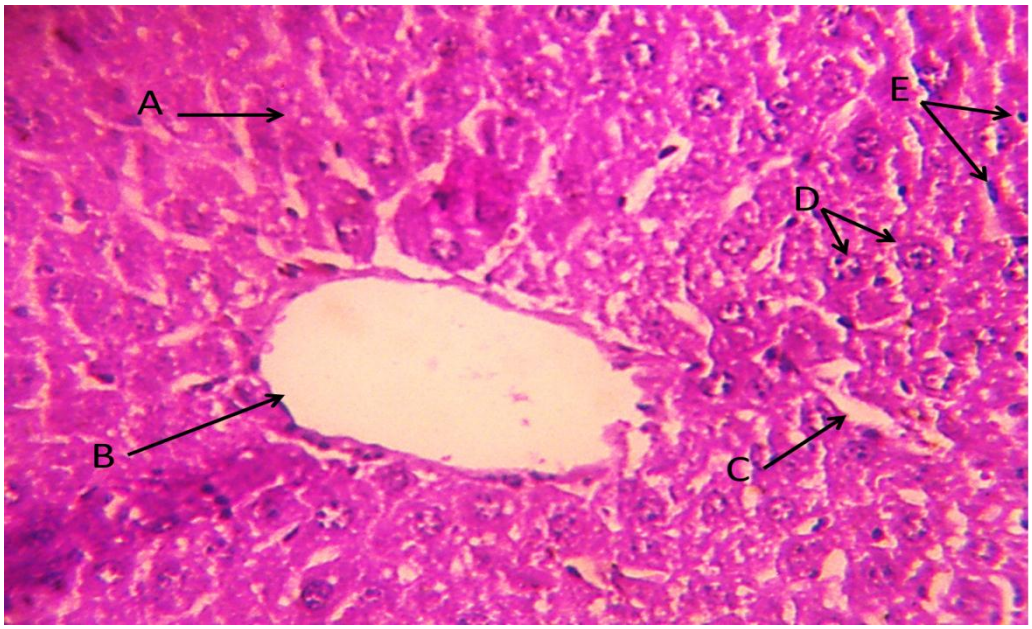


Fig (4-30): Transverse section of liver in control group which showed A- Hepatic cord, B- Central vein, C- Sinsoid, D- Hepatocyte, E- Kupffer cell. **H&E** stain 40X.

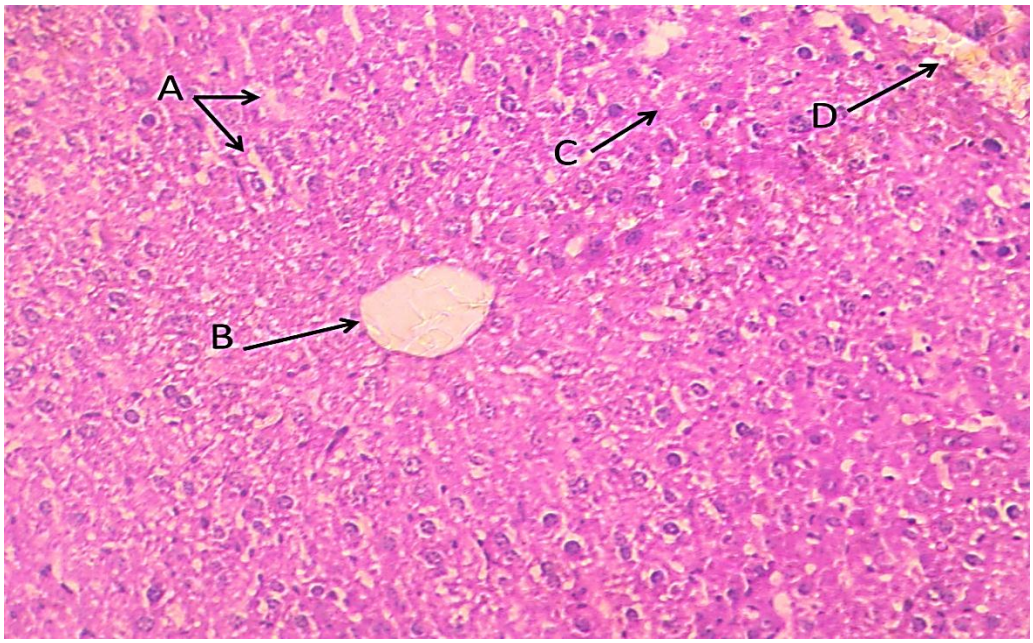


Fig (4-31): Transverse section of liver in control group which showed A- Sinusoid, B- Central vein, C- Blood vessel , D- Sinusoid. **H&E** stain 20X.

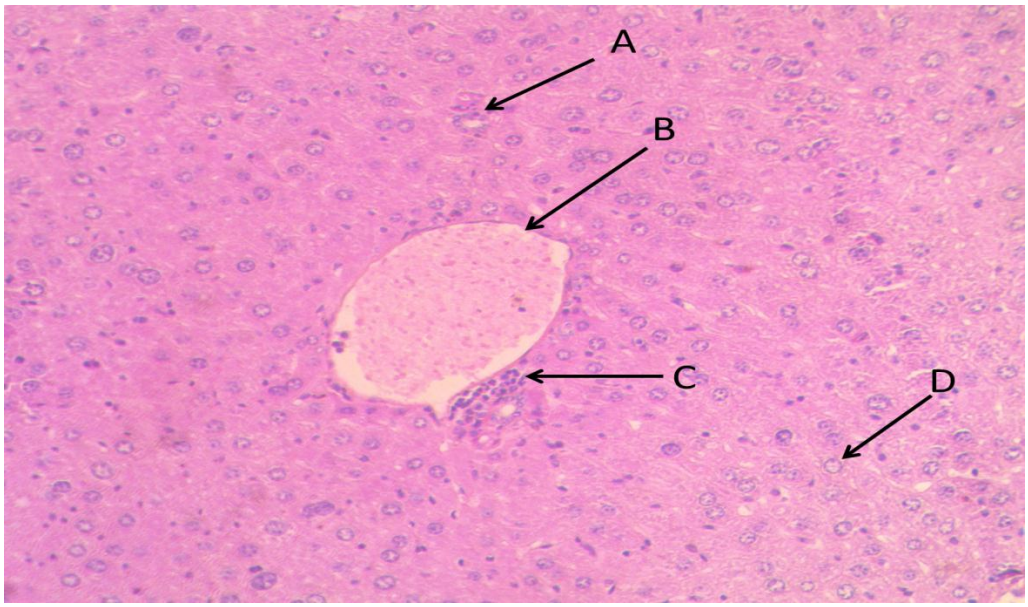


Fig (4-32): Transverse section of liver in treated group with *Nerium oleander* which showed A- Hepatocyte destruction , B- Blood congestion, C- Inflammatory cells, D- Cellular destruction. **H&E** stain 40X.

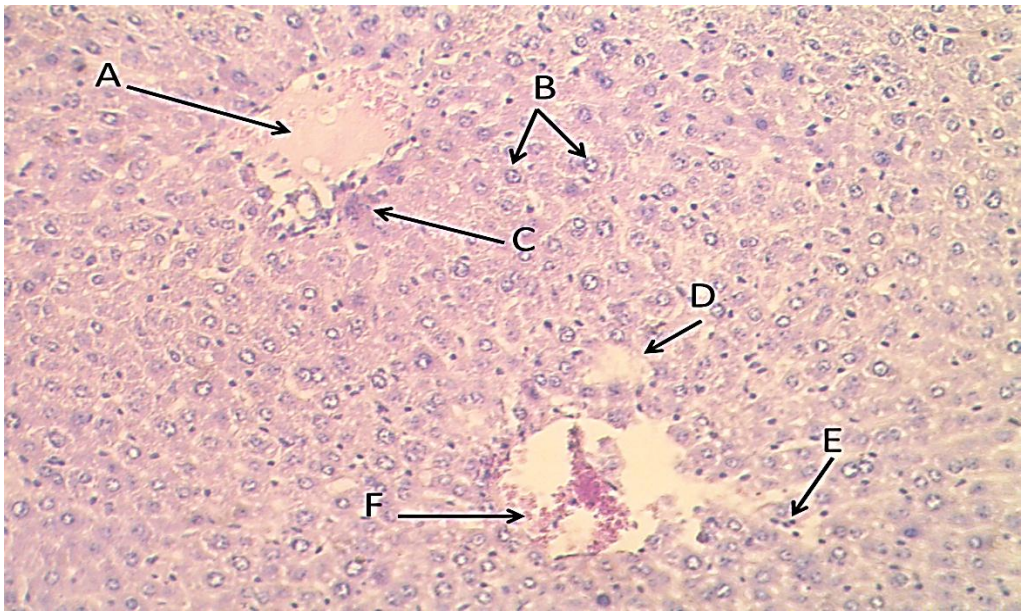


Fig (4-33): Transverse section of liver in treated group with *Nerium oleander* which showed A- Necrosis, B- Hepatocyte, C- Inflammatory cells, D- Necrosis, E- Inflammatory cell , F- Central vein with congestion. **H&E** stain 40X.

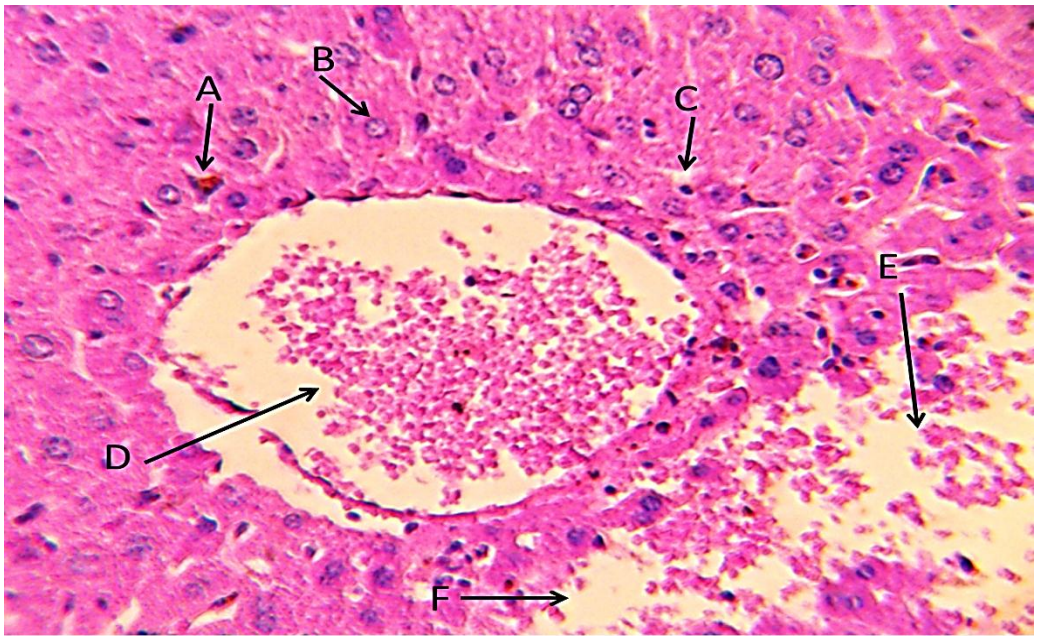


Fig (4-34): Transverse section of liver in treated group with *Nerium oleander* which showed A- Inflammatory cell , B- Hepatocyte, C- Sinusoid, D- Congested central vein, E- Hemorrhage, F- Necrosis. **H&E** stain 40X.

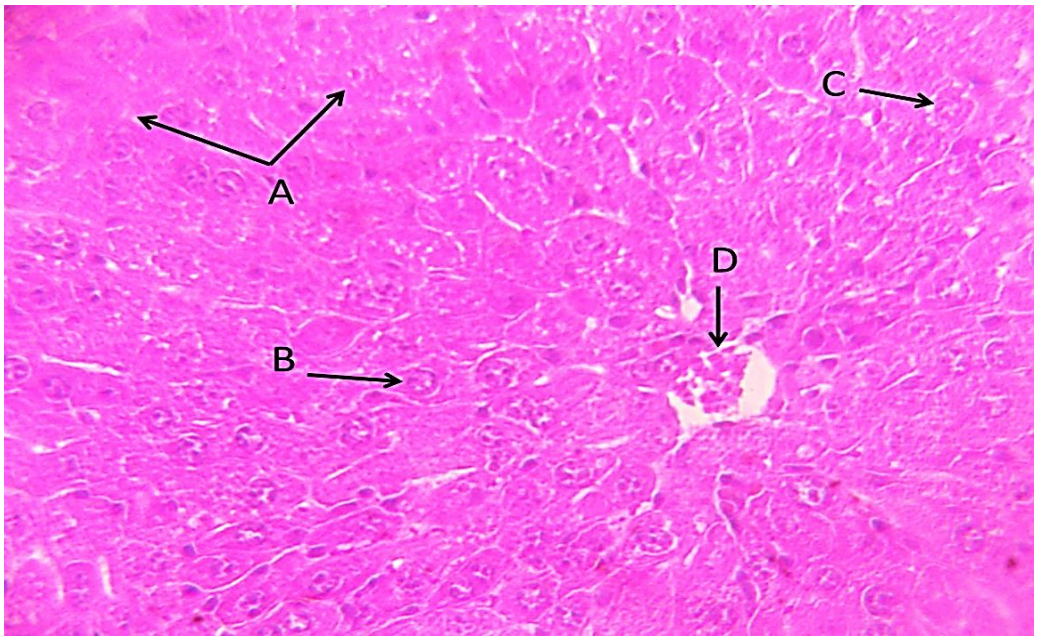


Fig (4-35): Transverse section of liver in treated group with lead which showed A- Prominent degeneration, B- Hepatocyte with abnormal nuclei, C- Degeneration in hepatocyte, D- Central vein filled with blood. **H&E** stain 40X.

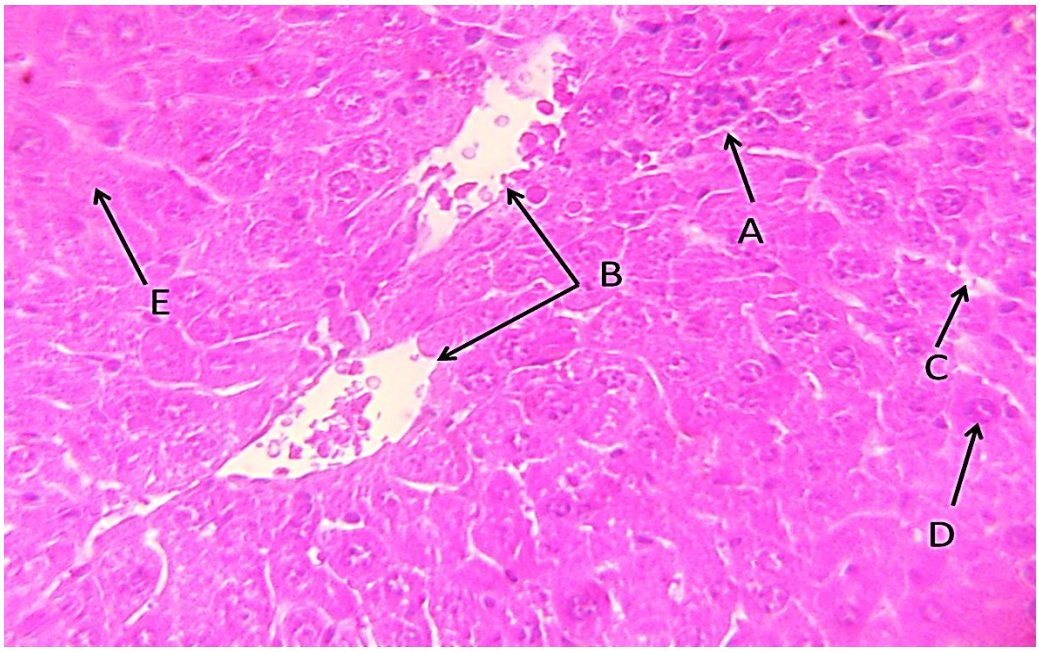


Fig (4-36): Transverse section of liver in treated group with lead which showed A- Inflammatory cells, B- Congestion, C- Necrosis, D- Hypertrophy in hepatocyte, E- Necrosis. **H&E** stain 40X.

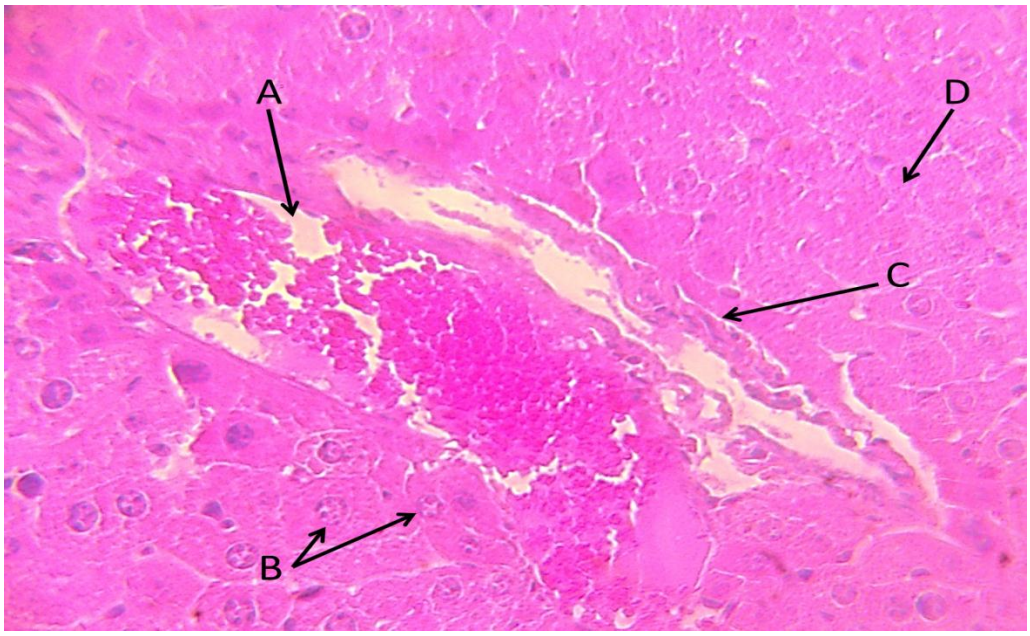


Fig (4-37): Transverse section of liver in treated group with lead which showed A- Central vein with congestion, B- Swelling in hepatocytes, C- Inflammatory cells, D- Necrosis. **H&E** stain 40X.

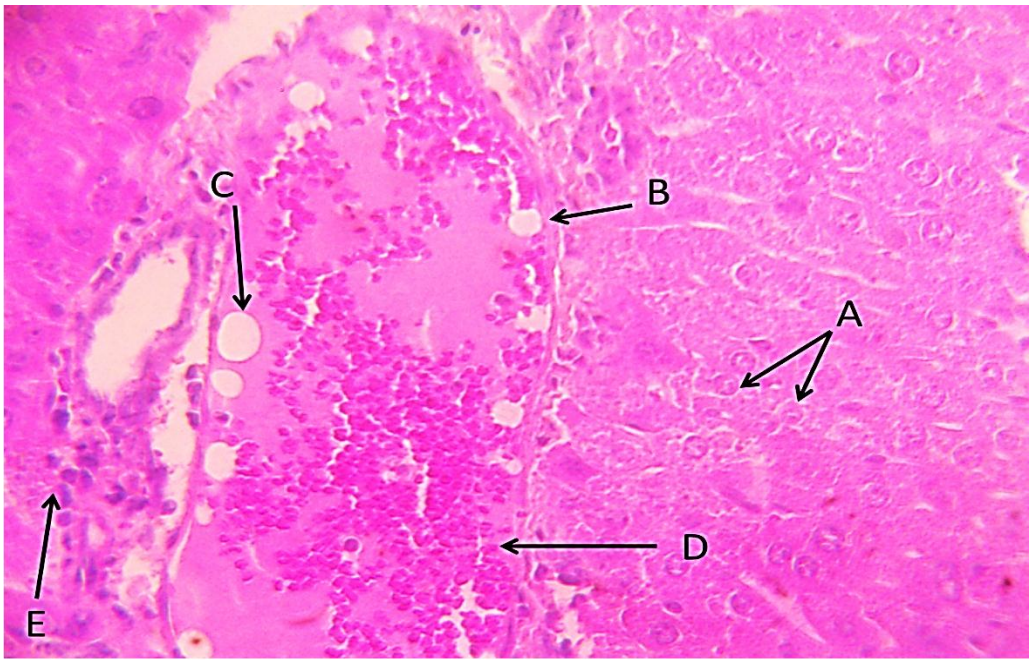


Fig (4-38): Transverse section of liver in treated group with lead which showed A- Hepatocyte without nuclei, B- Degeneration, C- Empty vacuole , D- Hemorrhage, E- Inflammatory cells. **H&E** stain 40X.

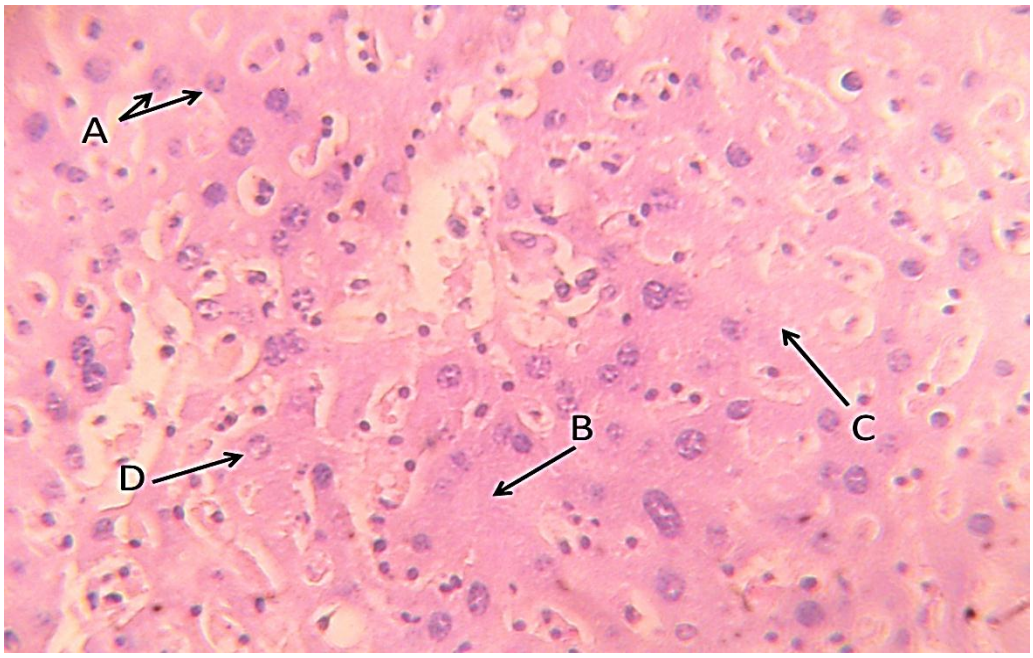


Fig (4-39): Transverse section of liver in treated group with Compound which showed A- Necrosis in hepatocyte, B- Necrosis in parenchyma, C- Necrosis in tissue, D- Irregular hepatic cord (plate). **H&E** stain 40X.

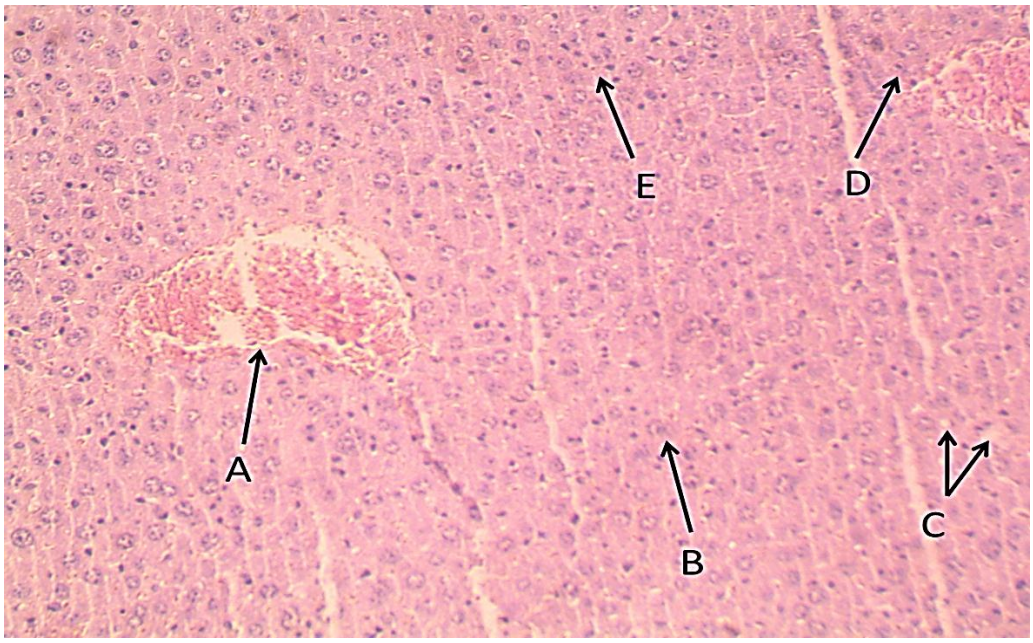


Fig (4-40): Transverse section of liver in treated group with Compound which showed A- Central vein congestion, B- Hepatocyte, C- Necrosis, D- Inflammatory cell, E- Kupffer cell. **H&E** stain 20X.

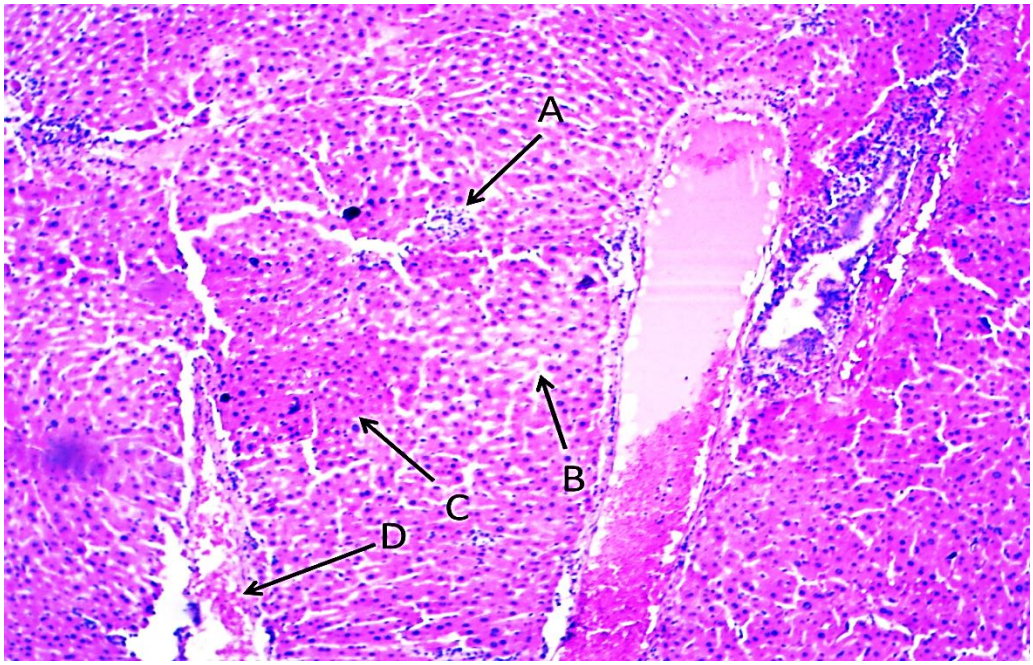


Fig (4-41): Transverse section of liver in treated group with Compound which showed A- Inflammatory cell, B- Irregular sinusoid, C- Abnormal cellular proliferation, D-Congestion. **H&E** stain 40X.

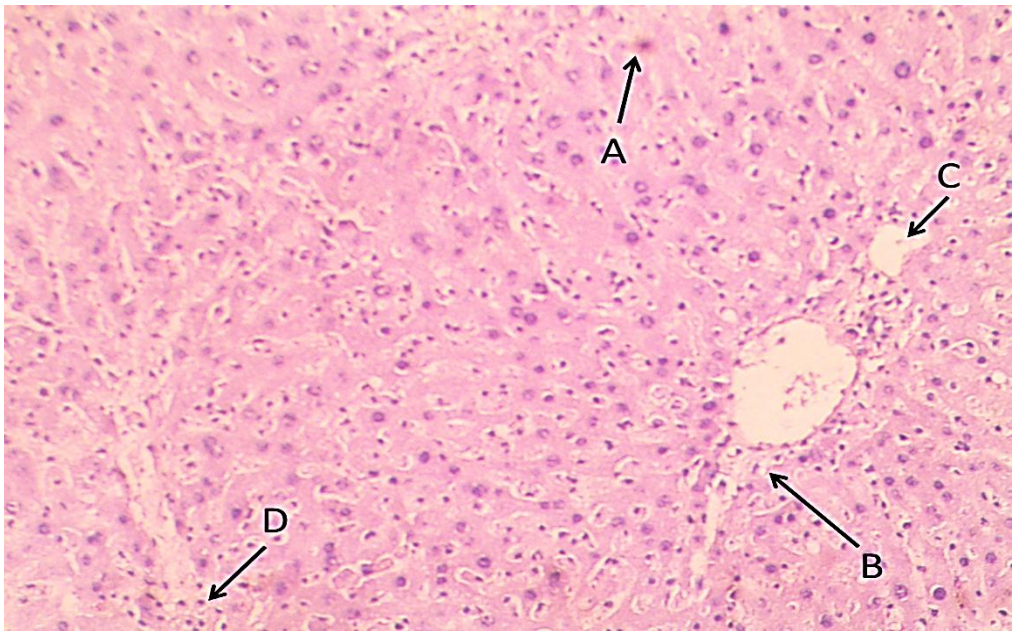


Fig (4-42): Transverse section of liver in treated group with Compound which showed A- Necrosis, B-Kupffer cell, C- Degeneration, D- Inflammatory cell. **H&E** stain 40X.

4.1.4: The Histological Results of Kidney

4.1.4.1: The Histological Results of Kidney of Control Group

The present study showed the diameter of renal corpuscle in the control group was (16.35±0.350µm),(Table 4-3).The current results showed that the kidney was surrounded by thin connective tissue capsule. The cortical region of kidney have renal corpuscle in normal structure, which composed of glomerular capillaries surrounded by double layer of simple squamous epithelium, parietal and visceral layer called Bowman's capsule and limited between two layers the space called Bowman's space (Fig.4-43).

The current result showed the renal tubules in the cortical region near the renal corpuscle, the renal tubules which have very narrow lumen and lined by high cuboidal epithelia called proximal convoluted tubules, the proximal convoluted tubule was (2.54±0.114µm) in diameter (Table 4-3). The brush border on the apical portion of epithelial cells which protruded inside the lumen of proximal convoluted tubule (Fig.4-44).

The wall of proximal convoluted tubule have limited cells that composed of the proximal convoluted tubules. The cell have oval or elongated nuclei centrally location. The tissue section of kidney in the control group showed that the other renal tubule called distal convoluted tubules which have very wide lumen compared with proximal convoluted tubules in same group (Fig.4-44).

The distal convoluted tubules was (5.09±0.129µm) in diameter showed in (Table 4-3). The current results showed the distal convoluted tubule were lined by very low simple cuboidal epithelia without brush border.

The descending branch of Henle loop have narrow lumen with (2.75±0.102µm) in diameter (Table 4-3), the inner surface lined by simple squamous epithelia (Fig.4-45).

The result showed the ascending branch of Henle loop was wider lumen than the lumen of descending branch with (5.23±0.091µm) in diameter (Table 4-3), the lumen lined by simple squamous epithelia.

The tissue section of kidney showed the descending and ascending limp of Henle loop in normal structure. The collecting tubules were (7.43±0.122µm) in diameter (Table 4-3), and lined by cuboidal epithelium. Two types of cells compose the

epithelium of collecting ducts which included the principle cells (light cells) and intercalated cells (dark cells) with dark and pale nuclei.

4.1.4.2: The Histological Results of Kidney Treated Group with *Nerium oleander* after 30 days orally administration

The tissue section of kidney showed that the renal corpuscle have significant increased in diameter was (13.05±0.320µm),(Table 4-3) which have significant decreased compared with control group.

The histological results of kidney in treated mice with *Nerium oleander* noted acute affected in tissue structures of kidney, the histological diagnosis included the kidney parenchyma have prominent tissue destruction exactly in the cortical region. (Fig.4-46). The histological change in the tissue section of kidney may be due to the *Nerium oleander* toxicity that lead to thus alterations, this result confirmed with (Adam *et al*., 2001) which showed the kidney of sheep and goats when treated with *Nerium oleander* caused destruction.

The result showed significant decreased in diameter of proximal convoluted tubules (4.73±0.127µm),(Table 4-3) compared with the control group. The tissue section of kidney have acute degeneration in the wall of proximal convoluted tubule and distal convoluted tubules, so, showed irregular wide spaces filled with blood between the proximal and distal convoluted tubules (Fig.4-46), the distal convoluted tubules have significantly decreased in diameter with (11.00±0.327µm),(Table 4-3) compared with the control group. The tissue section noted inflammatory cells aggregation in different locations of kidney, so, the results appeared many necrosis lesions in the cortical region of treated kidney (Fig.4-47), this result may be due to the oleandrin toxicity which caused accumulation of large amounts of toxin lead to severe destruction in proximal convoluted tubule and distal convoluted tubules and blood congestion was result agreement with (Ni *et al*., 2002) which showed the kidney of goats after exposure to *Nerium oleander* caused degeneration in the kidney tissue and tubules.

The descending branche of Henle loop was (3.89±0.093µm) in diameter (Table 4-3). While the ascending branch of Henle loop was (9.18±0.288aµm) in diameter (Table 4-3). Both diameters of descending and ascending branches of the Henley

loop have significantly increased compared with the control group. This increased in diameter of Henle loop branches may be because increased infiltrate of the toxic oleander substance.

The tissue section of kidney after treated with aqueous extracted of *Nerium oleander* noted many renal corpuscle progressive and appeared as dark spots, on the other hand some renal corpuscle have glomerular capillaries with prominent lumen, that congestion with blood, the mesangial cells between the glomerular capillary have dark oval nuclei, the watery extract of *Nerium oleander* have prominent effect in reduced the width of Bowman's space in most renal corpuscle, the Bowman's space in the most renal corpuscle appeared as clear narrow space the glomerular capillaries in the renal corpuscle (Fig.4-46,48). This histological result was may be due high stress on nuclei or destruction in the junction between renal tubules. This histological result coincide with (Adams, 1995) which observed the kidney after exposure to *Nerium oleander* caused degenerative in renal corpuscle and glomerular components. The diameters of collecting duct (11.41±0.163µm),(Table 4-3) have significantly increased compared with control group.

The histological results in treated kidney showed prominent destruction in glomerular capillaries in renal corpuscle, the Bowman's capsule have prominent parietal layer, while the visceral layer of Bowman's capsule wasn't clear (Fig.4-50). This result confirmed with (Adam *et al*., 2001) which noted the oleandrin toxicity caused injury in glomerular capillaries, destruction in the cells and Bowman's capsule.

The histological results of the kidney after treated with *Nerium oleander* showed most of the renal tubules were distinguished by didn't have a true wall, the disappearance of the cells that responsible for the wall structure of the proximal convoluted tubules. The proximal convoluted tubules have very wide lumen without brush border, the internal lumen of the proximal convoluted tubules lined by cuboidal epithelia, the deceased of the hight of the epithelia in the proximal convoluted tubules was because of the increased the amount of infiltration inside the proximal convoluted tubules that lead to pressing on the apical surface of the epithelial layer (Fig.4-49).

This result me be due to the effects of oleander glycosides on the kidney of mice caused damage in cortical region. This histological change agreement with (Baselt, 2017).

which showed necrosis in renal tubules and damage in epithelial tissue of the tubules.

4.1.4.3: The Histological Results of Kidney Treated Group with Lead after 30 days orally administration

The histological result of kidney after treated time noted the renal corpuscle have significant decreased in diameter was (13.76±0.294µm), (Table 4-3) when compared with renal corpuscle in the previous treated and control group.

The renal corpuscle have abnormal glomerular capillaries and Bowman's capsule, the tissue section showed prominent destruction in the glomerular capillaries, the other capillaries were congested with blood, the mesangial cells have irregular dark nuclei, no, prominent normal Bowman's space, the Bowman's capsule didn't have prominent structures (Fig.4-51).

This histological result agreement with (Rahymah *et al*., 2011) which noted the kidney when exposure to lead caused injury in renal tubules and renal corpuscle, defect in Bowman's capsule.

The diameter of proximal convoluted tubule (P.C.T.) was (6.66±0.182µm),(Table 4-3) which have significant increased compared with control group. The diameter of diameter distal convoluted tubule (D.C.T) was (12.27±0.293µm),(Table 4-3) showed significantly increased compared with control group. The tissue section of kidney showed the renal corpuscle surrounded by segmented Bowman's capsule, the result noted the Bowman's space filled with infiltrate or secretion, so noted many spherical vacuoles in Bowman's space (Fig.4-52). This histological change may be due to increase the rate of execration or elimination the toxic material through the kidney which caused vacuoles in and secretion in Bowman's capsule. This result agreement with (Altaee, 2011) which noted the increase infiltrate of toxic material caused damage in renal corpuscle and Bowman's space. The histological result showed the wall of proximal convoluted have prominent degeneration, the wall of proximal convoluted tubule losts of cells that composed of its wall. Most proximal convoluted tubule didn't have prominent simple cuboidal epithelia that lining the inner surface of tubules (Fig.4-52). These results may be due the accumulation of free radicals of lead in cortical region of kidney, was result confirmed with (Al-Farwachi *et al*.,

2008) which noted the kidney of mice after treated with lead, accumulation of free radical in in kidney tissue caused degenerative in the wall of P.C.T and cells of tubules.

The tissue section of proximal convoluted tubule showed didn't have a brush border in the apical portion of epithelial cells, the proximal convoluted tubule have prominent wide lumen filled with thick secretion, the distal convoluted tubules (D.C.T.) didn't have normal wall, most distal convoluted tubules lost their cells that from its wall and have prominent destruction, the tissue section showed isolated the epithelial layer in distal convoluted tubules and accumulation in the lumen of distal convoluted tubules (Fig.4-53,54), the lumen of distal convoluted tubules was filled with secretion. This histological change may be due to increased infiltration caused pressure on wall of distal convoluted tubules and the proximal convoluted tubule. Were result similar to (Altaee, 2011) which showed the kidney of mice after treated with lead caused damage in the brush border of the proximal convoluted tubule and cell wall of the distal convoluted tubules

The histological result showed prominent blood congestion and blood hemorrhage in some location between the renal tubules, the result showed abnormal cellular aggregation between renal tubules, so, the result noted the inflammatory cells as small clusters around the abnormal renal tubules (Fig.4-55). This histological change may be due to hydrostatic pressure on the blood vessels and renal tubules caused blood congestion and blood hemorrhage, were result coincide with (Adiguzel and Kalender, 2015) which showed the lead caused injury in renal tubules and blood vessels because increase in the pressure on these tubules and vessels.

The tissue section of kidney noted many wide empty cystic dilation in the medullary regions of kidney while other dilation filled with secretion (Fig.4-56). The tissue section showed that the descending branches of Henle loop have significant increased in diameter (4.32±0.112µm),(Table 4-3) compared with control group. The result showed significant increase in diameter of ascending branch of Henle loop became (8.22±0.216 µm),(Table 4-3) compared with the control group.

The tissue section noted the collecting ducts have (10.47±0.312µm) in diameter ,(Table 4-3) which have significant increased compared with control group.

The histological result of kidney after treated with lead noted abnormal Henle loop branches, the Ascending and descending branch of henle have abnormal wide lumen, both branches have prominent effects that lead to effects on the cells in the wall of

abnormal Henle loop branches, the Henle loop arms appeared filled with secretion. The histological result may due to materials toxicity or high toxicity of lead caused damage the wall of henle loop branches, was result agreement with (Al-Farwachi *et al*., 2008) which showed when the kidney is exposed to lead, it caused injury and degeneration in descending and ascending branch of Henle loop.

4.1.4.4: The Histological Results of Kidney Treated Group with (Lead+*Nerium oleander*) after 30 days orally administration

The histological results of kidney after treated with lead and *Nerium oleander* compound have abnormal renal corpuscle, the diameter of affected renal corpuscle was (14.82±0.228µm),(Table 4-3) which have significant decreased compared control group.

The kidney in the treated mice for thirty days of experimental time have histological changes in the tissue sections, the cortical region of kidney have prominent wide space, some of spaces filled with blood, the tissue section showed abnormal cellular proliferation in the cortex of kidney and aggregation of inflammatory cells located between abnormal renal corpuscles (Fig.4-57). This histological change may be due to the effect toxins on kidney tissue that lead to damage. This result was similar to (Mohammed *et al*., 2017) which showed destruction in cortical region, proliferation of inflammatory cells and blood congestion.

The diameter of proximal convoluted tubule (P.C.T.) was (4.56±0.112µm),(Table 4-3), the present result showed the distal convoluted tubules have wide lumen with (13.64±0.489µm) in diameter (Table 4-3), this two branches have significantly increased compared with control group. The histological results showed the renal corpuscles have prominent destruction in the glomerular capillaries, the mesangial cells have oval dark nuclei which isolated as small groups in the renal corpuscle, some of renal corpuscle have Bowman's capsule only with small aggregation of mesangial cells, the glomerular capillaries didn't have prominent lumen in the most tissue section (Fig.4-58). This histological result may be due to accumulation of lead in renal corpuscle and cells caused this damage and destruction in kidney tissue, were result confirmed with (Kaur *et al*., 2018) which showed the kidney after treated with lead caused degeneration in glomerular structure, defect in Bowman's capsule.

The tissue section showed the proximal convoluted tubules have prominent changes which have acute destruction in the epithelial layer which accumulation in the lumen of proximal convoluted tubule (Fig.4-59). This histological change confirmed with (Portier, 2012) which noted the kidney after treated with lead caused damage in renal tubules.

The proximal convoluted tubules didn't have normal structures in the tissue section of kidney after treated with lead and *Nerium oleander*. The cells that composed of the wall of proximal convoluted tubule were aggregation in the central lumen of proximal convoluted tubule, most cells loss their nuclei most the proximal convoluted tubule appeared loss the epithelial layer that lining the inner surface of renal tubules and no prominent brush border, the tissue section showed most proximal convoluted tubule have acute destruction in the histological structures of proximal convoluted tubule wall, so, aggregation of inflammatory cells exactly between affected renal tubules (Fig.4-60). These histological results may be because destruction in the junction between renal tubules caused degenerative in proximal convoluted tubule and. This result agreement with (Nordberg *et al*., 2009) which showed when the kidney is exposed to lead caused severe degeneration in proximal convoluted tubule, proliferation of inflammatory cells.

The tissue section showed that the descending branches of Henle loop have significant increased in diameter (3.36±0.081µm),(Table 4-3) compared with control group. The result showed significant increased in diameter of ascending branch of Henle loop became (6.23±0.157µm),(Table 4-3) compared with the control group.

The diameters of collecting duct was (14.02±0.181µm),(Table 4-3) which have significant increased compared with control group. The distal convoluted tubules didn't have normal tissue structure in the kidney section, the distal convoluted tubules didn't have normal wall, no prominent epithelial layer, the wall have clear destruction and the residual epithelial cells accumulation in the central space in the distal convoluted tubules, the tissue section showed completely separated epithelial layer from the wall of distal convoluted tubules (Fig.4-61). This histological change may be due to high hydrostatic pressure due to the lead, this result coincide with (Mohammed *et al*., 2017) which noted necrosis in distal convoluted tubules and destruction in epithelial layer.

The Ascending and Descending branches of Henle loop have prominent lumen, the outer surface was normally, the internal lumen lined by simple squamous epithelial, the epithelial layer was isolated from the wall of Henle loop.

The Ascending branch of Henle loop filled with thick fluid in lumen of most Ascending Arms of Henle loop, the tissue section showed abnormal cellular proliferation between both branch of Hnele loop (Fig.4-62). This histological result agreement with (Kaur *et al*., 2018) which noted the kidney after treated with degenerative in Henle loop branches and Ascending branches filled with fluid.

Table (4-3): Measurement of kidney parts in study group in mice M± S.D

Treatment		Control	Lead	*Nerium oleander*	Compound
Diameter	Renal corpuscle	16.35±0.350[a]	13.76±0.294[c]	13.05±0.320[b]	14.82±0.228[b]
	D.C.T	5.09±0.129[d]	12.27±0.293[b]	11.00±0.327[c]	13.64±0.489[a]
	P.C.T	2.54±0.114[c]	6.66±0.182[a]	4.73±0.127[b]	4.56±0.112[b]
	A.B	5.23±0.091[d]	8.22±0.216[b]	9.18±0.288a	6.23±0.157[c]
	D.B	2.75±0.102[d]	4.32±0.112[a]	3.89±0.093[b]	3.36±0.081[c]
	C.D	7.43±0.122[d]	10.47±0.312[c]	11.41±0.163[b]	14.02±0.181[a]

Different letters vertically indicate the existence of significant differences between the averages at the possibility of (0.01).

(Table4.3-1) Results of ANOVA analysis of renal corpuscle of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
Renal corpuscle	Between Groups	308.729	3	102.910	22.601	.000
	Within groups	892.464	196	4.553		
	Total	1201.193	199			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Nerium oleander	50	13.0596		
Lead	50	13.7628		
Compound	50		14.8228	
Control	50			16.3580
Sig.		.101	1.000	1.000

(Table4.3-2) Results of ANOVA analysis of distal convoluted tubules of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
DCT	Between Groups	2126.103	3	708.701	126.140	.000
	Within groups	1101.201	196	5.618		
	Total	3227.303	199			

Duncan

TRT	N	Subset for alpha = 0.01			
		1	2	3	4
Control	50	5.0926			
Nerium oleander	50		11.0040		
Lead	50			12.2726	
Compound	50				13.6442
Sig.		1.000	1.000	1.000	1.000

(Table4.3-3) Results of ANOVA analysis of proximal convoluted tubules of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
PCT	Between Groups	426.159	3	142.053	151.253	.000
	Within groups	184.078	196	.939		
	Total	610.237	199			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Control	50	2.5426		
Compound	50		4.5660	
Nerium oleander	50		4.7304	
Lead	50			6.6676
Sig.		1.000	.397	1.000

(Table4.3-4) Results of ANOVA analysis of ascending branch of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
A.B	Between Groups	490.443	3	163.481	79.957	.000
	Within groups	400.745	196	2.045		
	Total	891.188	199			

Duncan

TRT	N	Subset for alpha = 0.01			
		1	2	3	4
Control	50	5.2300			
Compound	50		6.2310		
Lead	50			8.2286	
Nerium oleander	50				9.1830
Sig.		1.000	1.000	1.000	1.000

(Table4.3-5) Results of ANOVA analysis of descending branch of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
D.B	Between Groups	68.619	3	22.873	47.262	.000
	Within groups	94.858	196	.484		
	Total	163.477	199			

Duncan

TRT	N	Subset for alpha = 0.01			
		1	2	3	4
Control	50	2.7578			
Compound	50		3.3638		
Nerium oleander	50			3.8996	
Lead	50				4.3200
Sig.		1.000	1.000	1.000	1.000

(Table4.3-6) Results of ANOVA analysis of collecting duct of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
Collecting duct	Between Groups	1109.082	3	369.694	171.612	.000
	Within groups	422.231	196	2.154		
	Total	1531.313	199			

Duncan

TRT	N	Subset for alpha = 0.01			
		1	2	3	4
Control	50	7.4350			
Compound	50		10.4762		
Nerium oleander	50			11.4124	
Lead	50				14.0224
Sig.		1.000	1.000	1.000	1.000

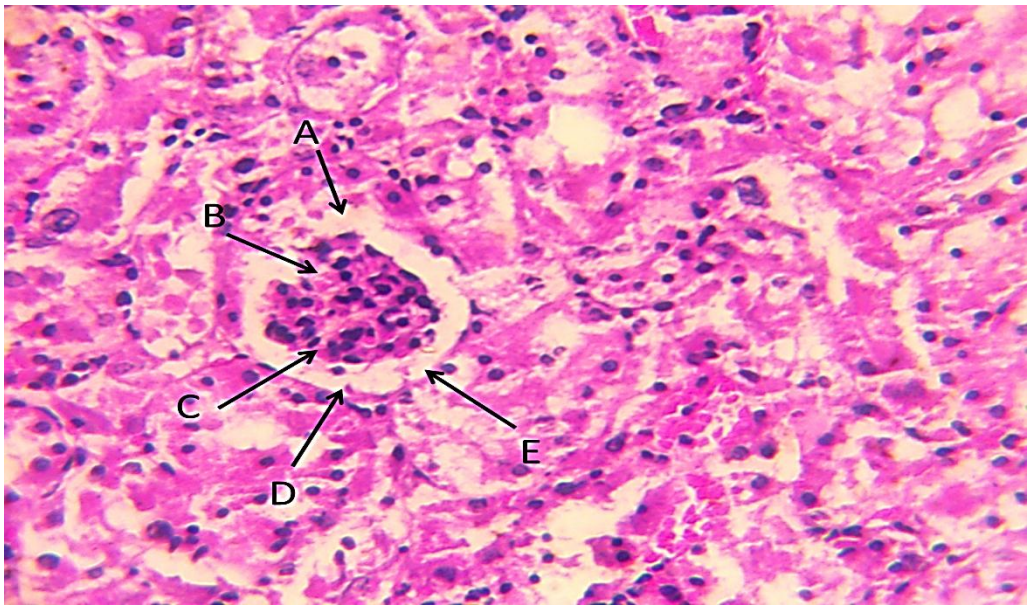


Fig (4-43): Transverse section of kidney in control group which showed A- Bowman space, B- Glomerular capillary, C- Visceral layer, D- Inter lobular Bowman capsule, E- Parietal layer. **H&E** stain 40X.

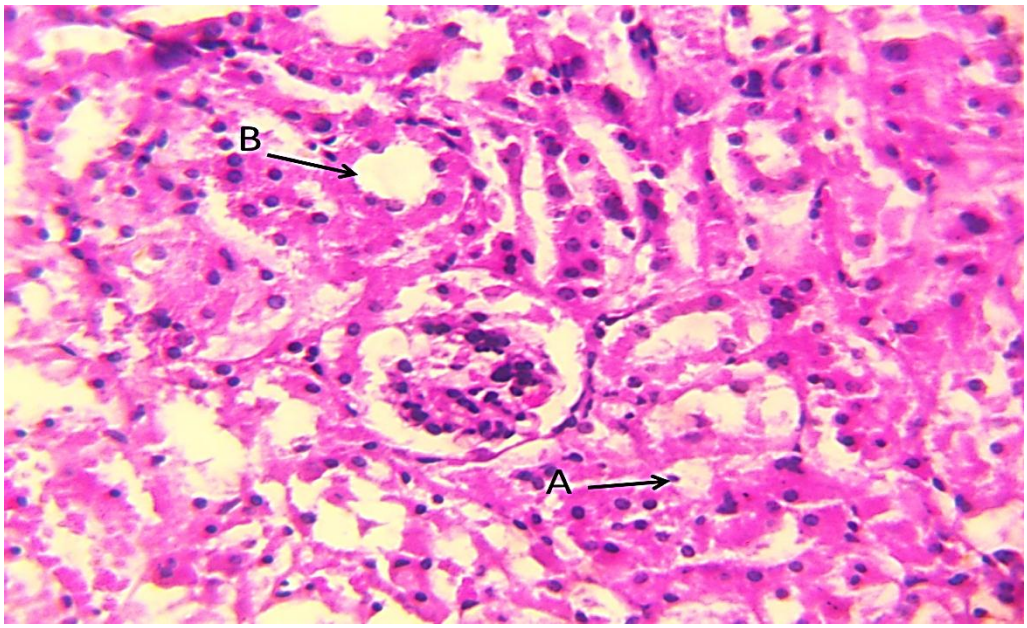


Fig (4-44): Transverse section of kidney in control group which showed A- Distal convoluted tubule, B- Proximal convoluted tubule . **H&E** stain 40X.

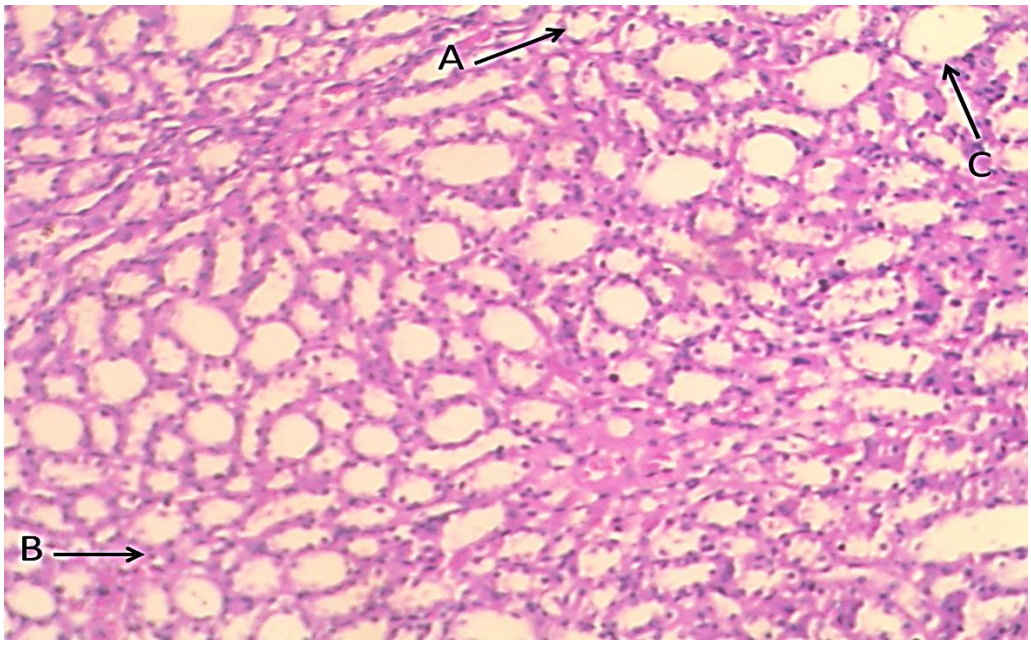


Fig (4-45): Transverse section of kidney in control group which showed A- Ascending branch of henle loop, B- Descending branch of henle loop, C- Collecting duct. **H&E** stain 20X.

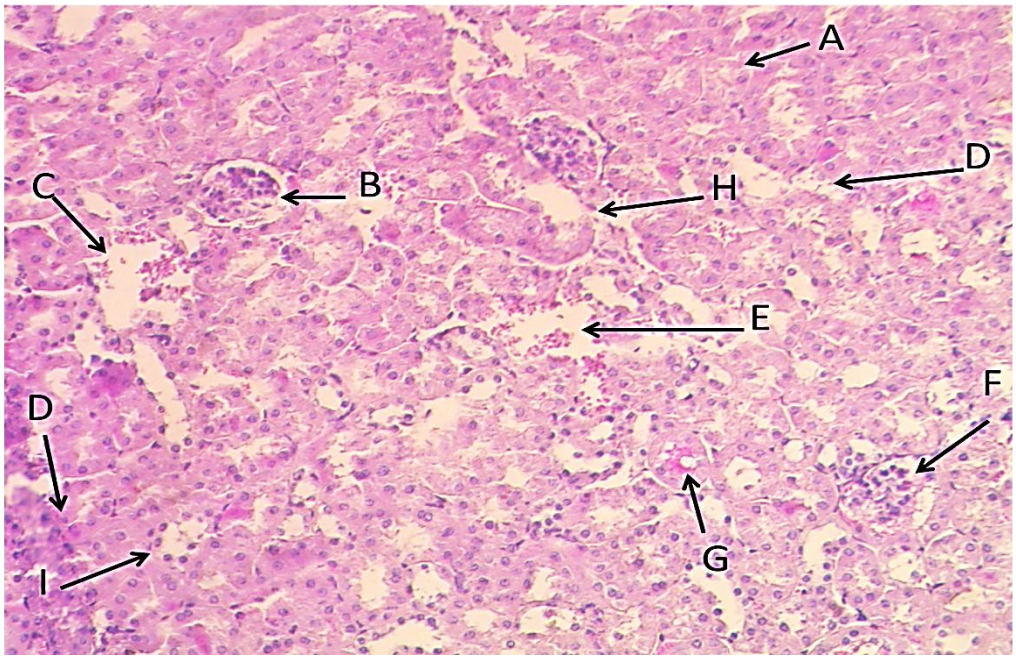


Fig (4-46): Transverse section of kidney in treated group with *Nerium oleander* which showed A- Proximal convoluted tubule, B- Glomerular capillary degeneration, C- Atrophy, D- Inflammatory cells, E-Desquamation, F- Necrosis, G- Thick inflammatory cells as cluster, I- Distal convoluted tubule. **H&E** stain 20X.

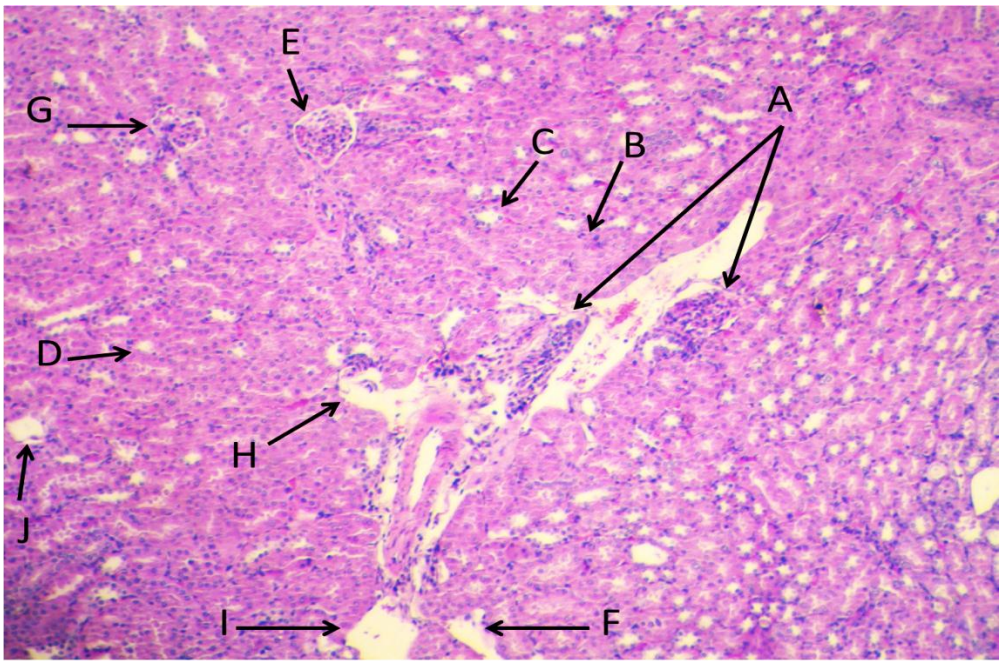


Fig (4-47): Transverse section of kidney in treated group with *Nerium oleander* which showed A- Cellular infiltration, B- Inflammatory cells, C- Distal convoluted tubule, D- Proximal convoluted tubule, E- Abnormal bowman space, F- Necrosis, G- Progressive renal corpuscle, H- Necrosis, I- Cystic dilation, J- Necrosis. **H&E** stain 20X.

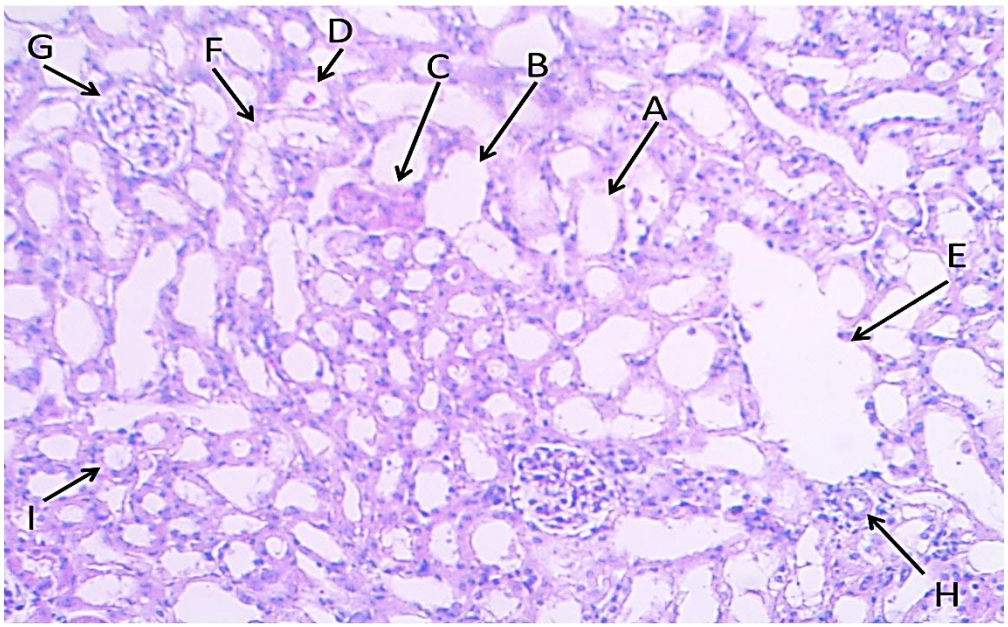


Fig (4-48): Transverse section of kidney in treated group with *Nerium oleander* which showed A- Necrosis, B- Wide D.C.T, C- Congestion, D- Wide P.C.T, E- Destruction of renal tubules, F- Vacuole, G- Very thin Bowman space, H- Blood vessel, I- Ascending branch of henle loop. **H&E** stain 40X

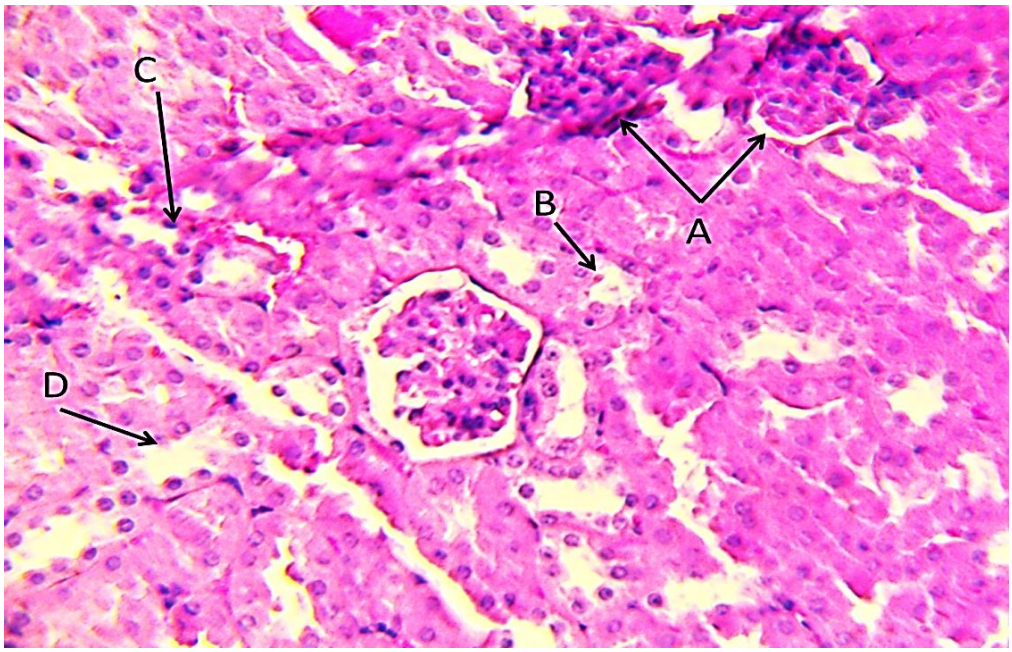


Fig (4-49): Transverse section of kidney in treated group with *Nerium oleander* which showed A- Atrophy, B- Prominent P.C.T, C- Cellular proliferation, D- Abnormal epithelia of D.C.T. **H&E** stain 20X

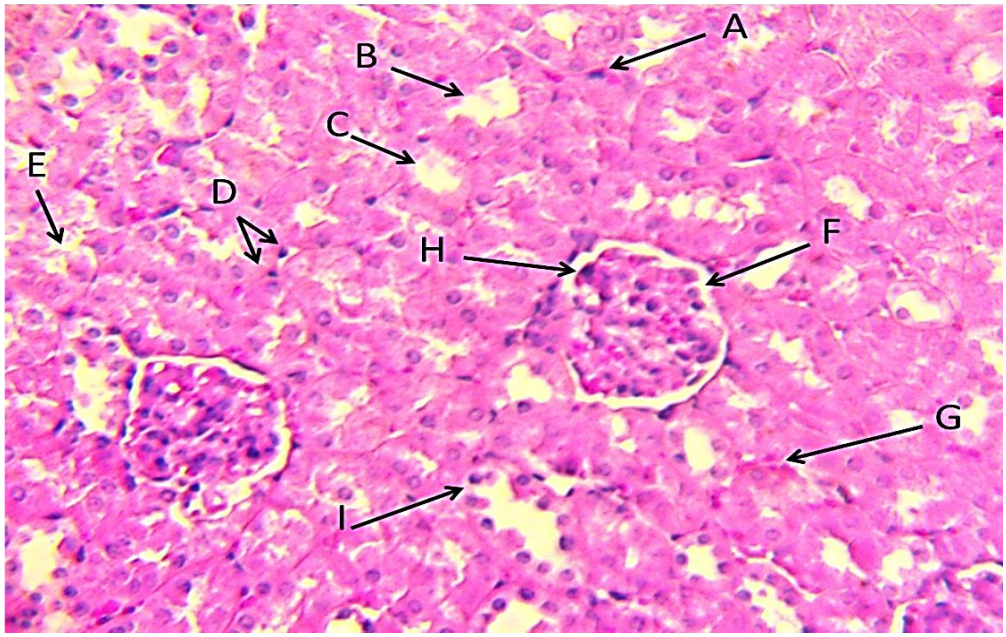


Fig (4-50): Transverse section of kidney in treated group with *Nerium oleander* which showed A- Inflammatory cell, B- Abnormal of wall P.C.T, C- Necrosis, D- Inflammatory cell, E-Sloughing epithelia tissue, F- Thin Bowman space, G- Blood hemorrhage, H- Bowman space, I- Sloughing epithelia tissue. **H&E** stain 20X.

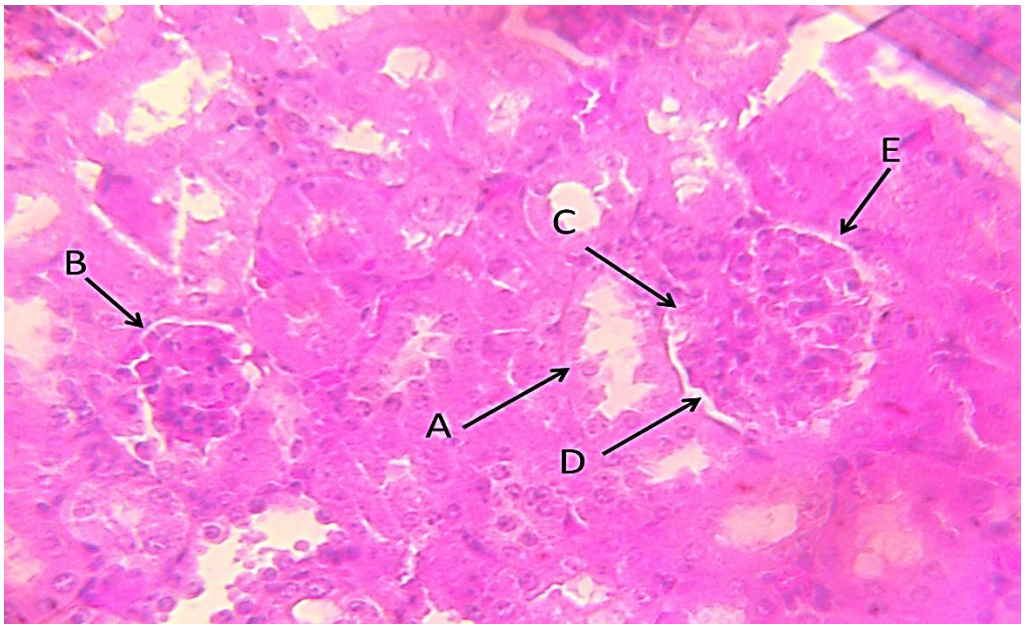


Fig (4-51): Transverse section of kidney in treated group with lead which showed A- Sloughing of epithelia in D.C.T. , B- Atrophy in renal corpuscle, C- Glomerular capillaries and other congested with blood, D- The bowmans capsule didn't have prominent structures, E- Parietal layer of bowmans capsule . **H&E** stain 20X.

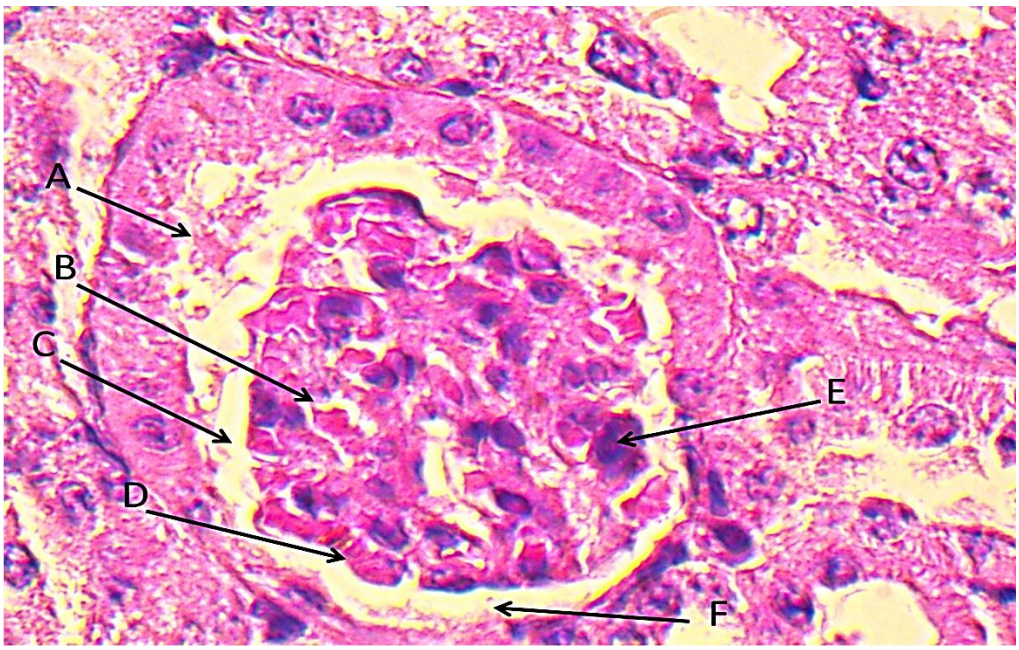


Fig (4-52): Transverse section of kidney in treated group with lead which showed A- Sloughing of epithelia in P.C.T, B- Glomerular capillary congestion with blood, C- Bowmans space, D- Visceral layer of B.C, E- Mesangial cell. **H&E** stain 40X.

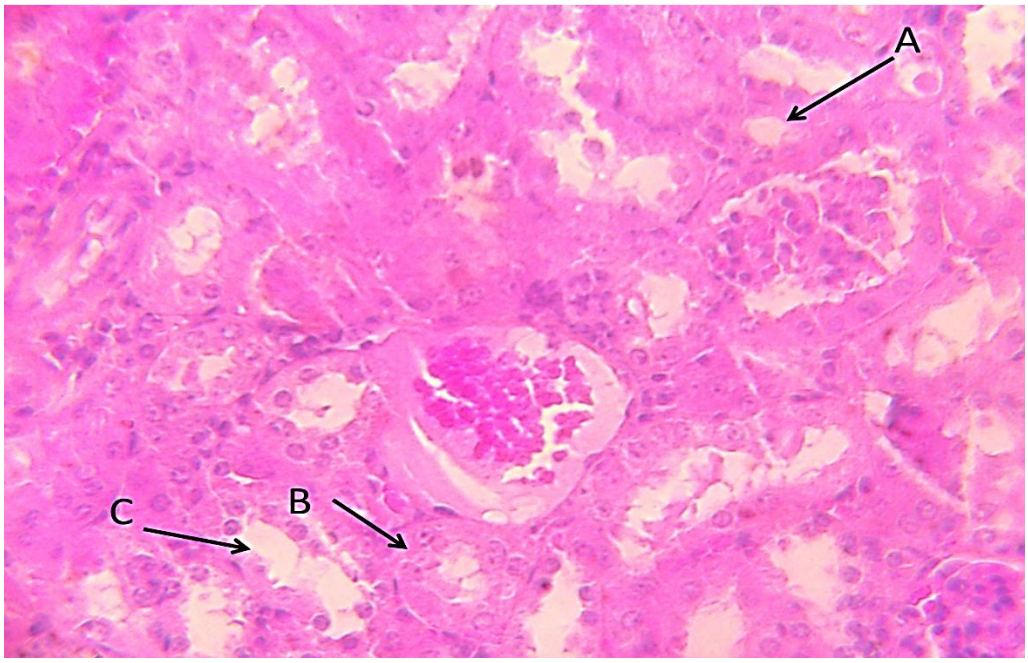


Fig (4-53): Transverse section of kidney in treated group with lead which showed A- P.C.T. have thick secretion, B- P.C.T have thick secretion , C- Wide lumen of D.C.T. **H&E** stain 20X.

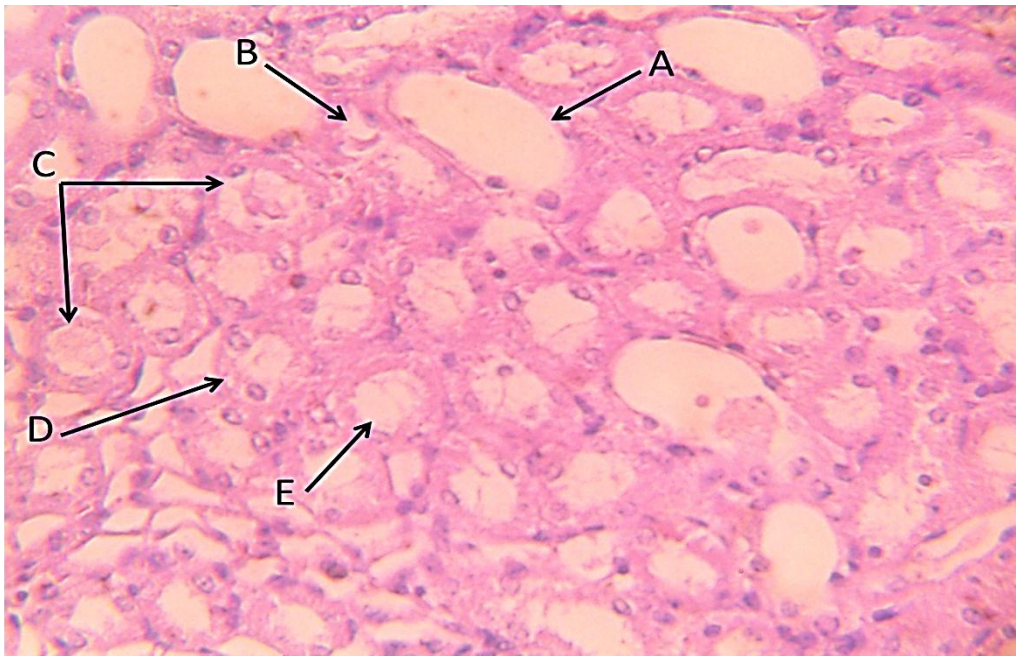


Fig (4-54): Transverse section of kidney in treated group with lead which showed A- Distal convoluted tubule, B- Necrosis , C- Thick infiltrate, D- Proximal convoluted tubule, E- P.C.T. necrosis. **H&E** stain 20X.

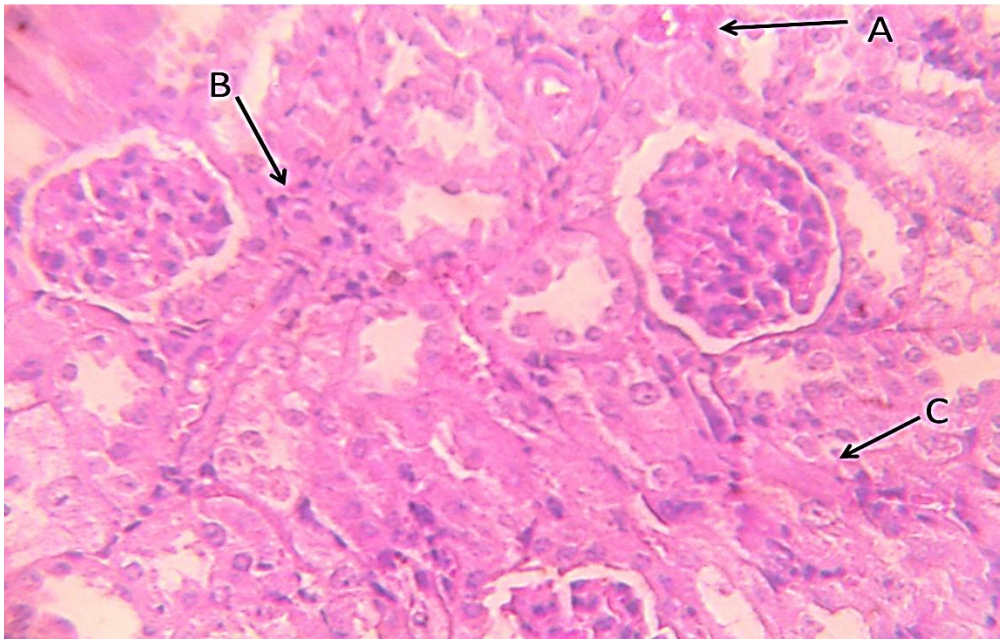


Fig (4-55): Transverse section of kidney in treated group with lead which showed A- Blood hemorrhage, B- Inflammatory cells, C- Blood congestion. **H&E** stain 40X.

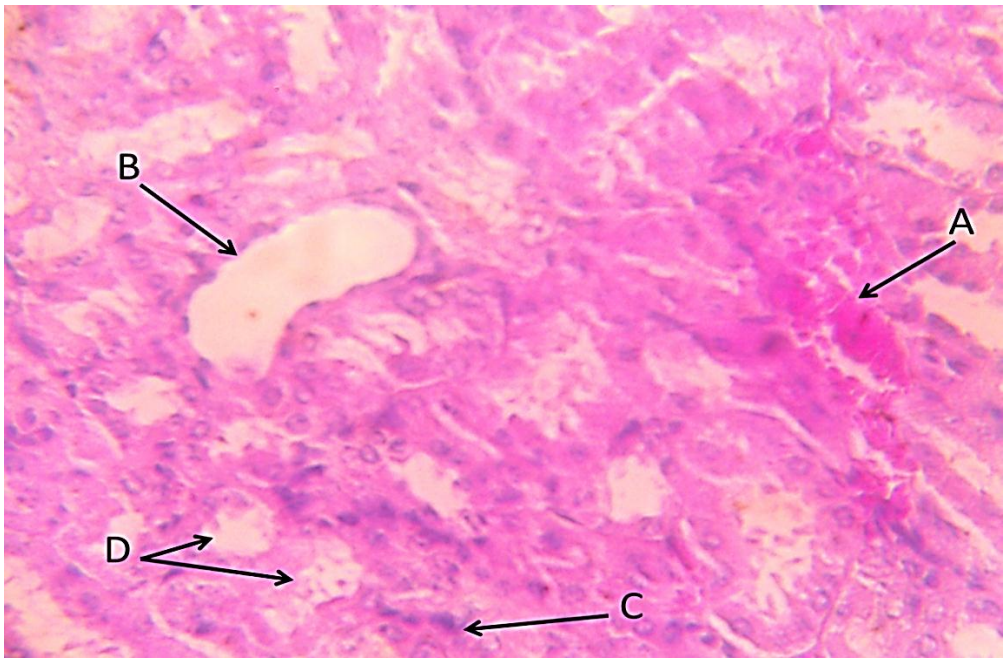


Fig (4-56): Transverse section of kidney in treated group with lead which showed A- Hemorrhage, B- Destruction of epithelia, C- Inflammatory cells aggregation, D- Acute degeneration. **H&E** stain 40X.

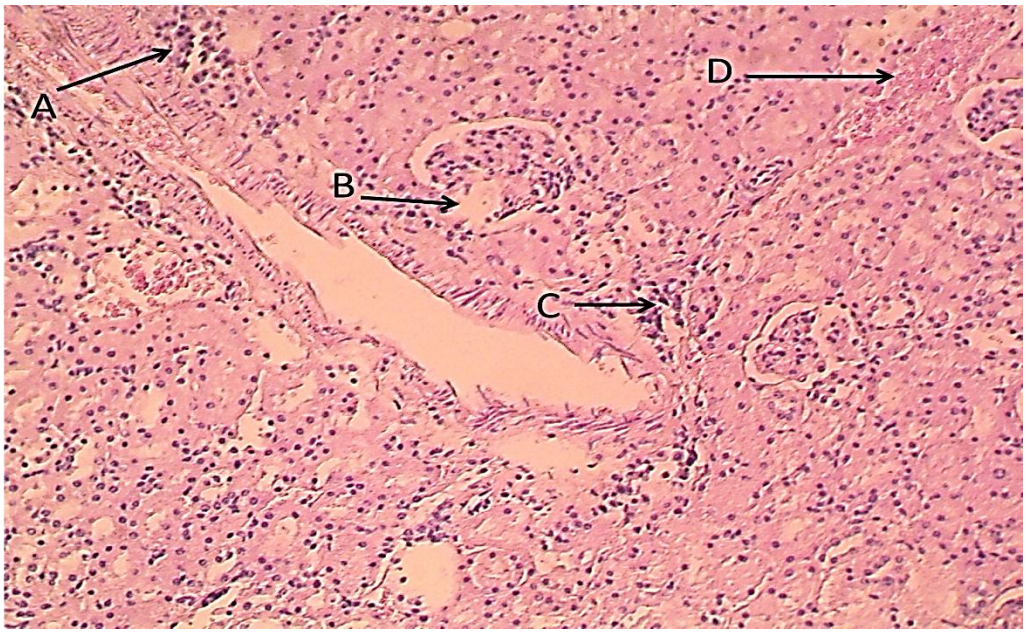


Fig (4-57): Transverse section of kidney in treated group with Compound which showed A- Aggregation of inflammatory cells, B- Degeneration, C- Cellular proliferation, D- Hemorrhage. **H&E** stain 20X.

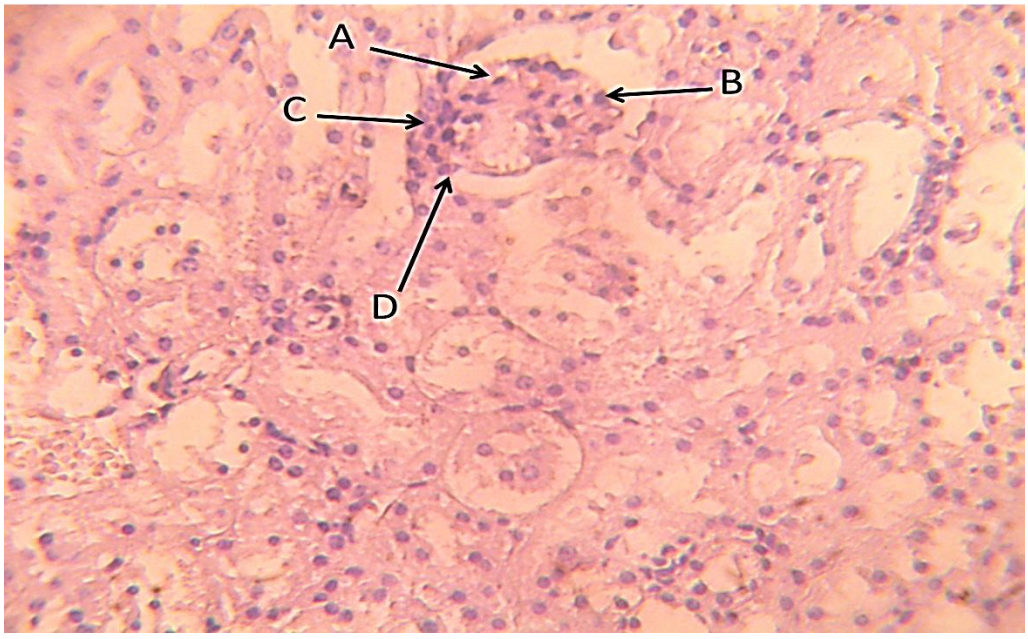


Fig (4-58): Transverse section of kidney in treated group with Compound which showed A- Atrophy in glomerular capillary , B- Glomerulus, C- Bowmans capsule, D- Renal corpuscle. **H&E** stain 20X.

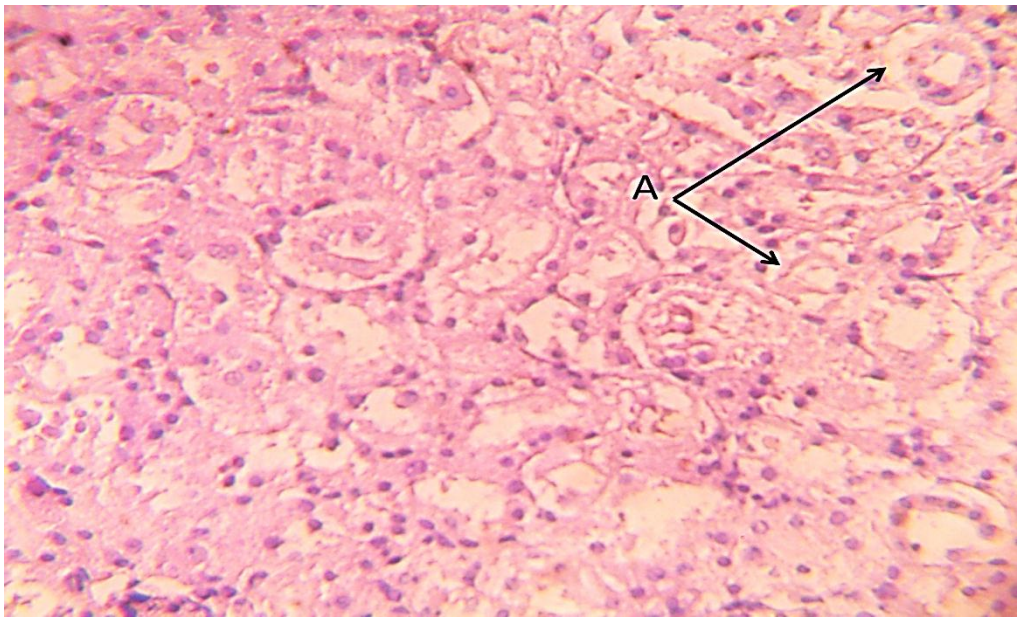


Fig (4-59): Transverse section of kidney in treated group with Compound which showed A- Destruction in epithelial layer of P.C.T. **H&E** stain 20X.

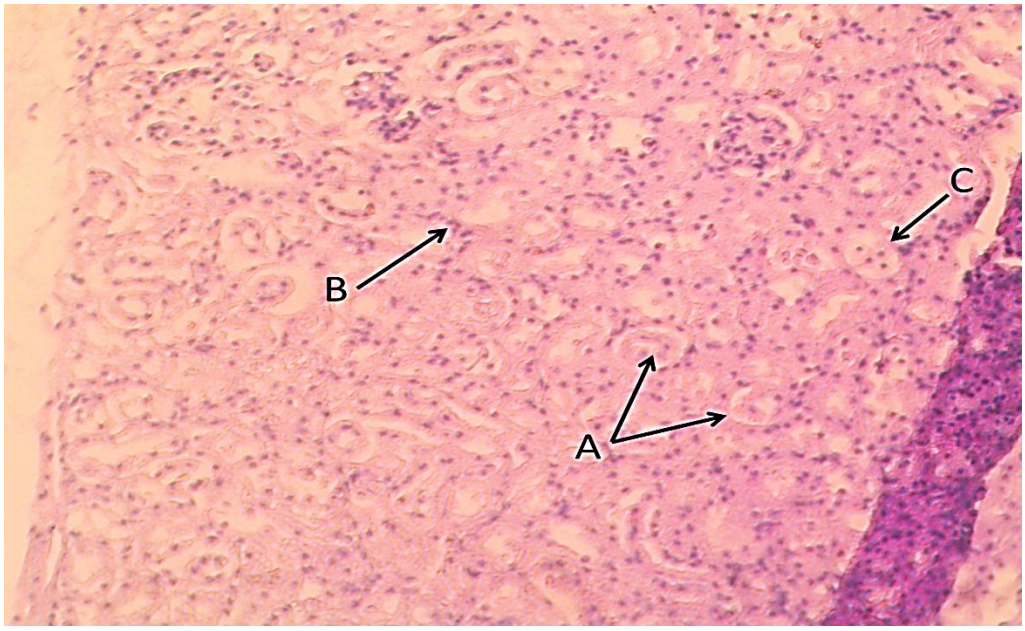


Fig (4-60): Transverse section of kidney in treated group with Compound which showed A- Destruction in the epithelial layer of P.C.T , B- Inflammatory cells, C- Destruction of epithelia in P.C.T. **H&E** stain 20X.

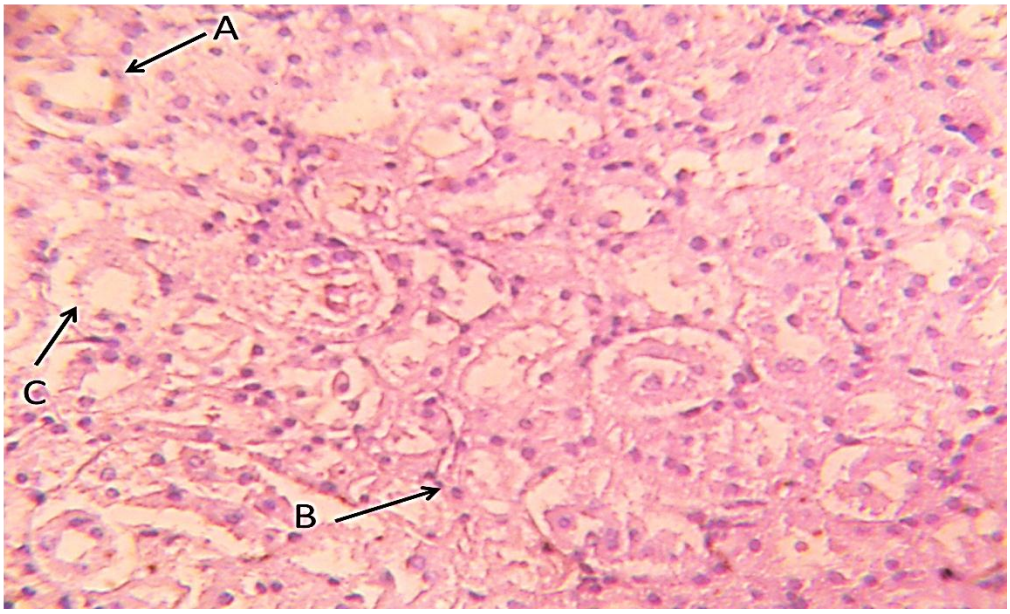


Fig (4-61): Transverse section of kidney in treated group with Compound which showed A- Epithelial cell accumulation in D.C.T, B- No wall in D.C. T, C- Separated epithelial layer from the wall of D.C.T.. **H&E** stain 20X.

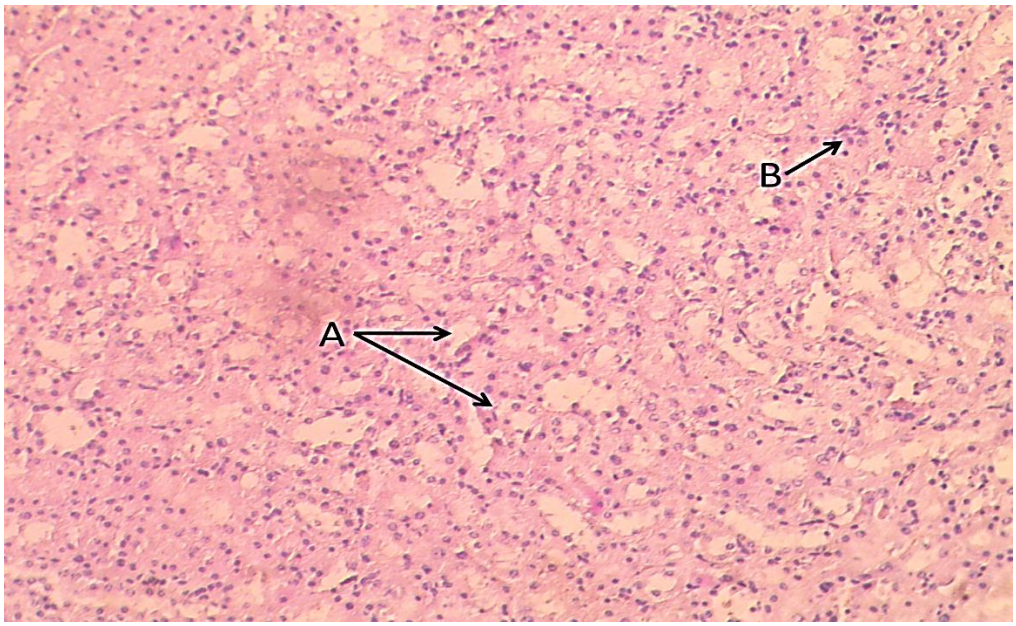


Fig (4-62): Transverse section of kidney in treated group with Compound which showed A- Destruction of epithelia in ascending branch of Henle loop, B- Inflammatory cells. **H&E** stain 10X.

4.2 The Biochemical results

4.2.1 The Biochemical Results of AST enzyme

Table (4-4) showed the level of AST enzyme in serum of control group was (365.15±12.295 U/L). The level of AST after treated with lead have significant increased (609.20±14.746 U/L) compared with control group. This result may be due to destruction of hepatocyte that lead to these changes. The destruction of hepatocytes lead to release high level of liver enzyme from destroid cells. This result confirmed with (Zhao *et al*., 2020) which showed significant increase in level of AST after treated with lead.

The level of AST after exposure to *Nerium oleander* (481.55±26.238 U/L) which didn't have significant increased compared with control group. This result may be due to oleandrin toxicity caused damage in liver tissue. These result not similar to (Altaee, 2011) which noted significant increased AST and ALT enzyme after oral administration by *Nerium oleander*. The level of AST serum after treated with compound (485.45±80.927 U/L) was significantly increase compared with control group.

4.2.2 The Biochemical Results of ALT enzyme

Table (4-4) showed the level of ALT in control group was (42.00±1.891U/L). The level of ALT enzyme after treated with lead was (143.35±10.957 U/L) which have significant increased compared with control group. This result may be due to accumulation of lead in liver caused damage in cytoplasm of hepatocyte. These result agreement with (Onyeneke and Omokaro, 2016) which showed significant increase in the AST, ALT and ALP level in mice after treated with lead.

The level of ALT after treated with *Nerium oleander* was (84.70±7.142 U/L) which have significant increased compared with control group. These result constant (Al-Farwachi *et al*., 2008) which noted significant increase activity of ALT and AST enzyme.

The level of ALT enzyme after treated with compound (216.20±20.666 U/L) was significantly increased compared with control group. The current explain high significantly increased compared with lead and *Nerium oleander*, this referred the treated group with compound is high toxicity effect on liver tissue caused release high level of enzyme.

4.2.3 The Biochemical Results of Urea

The present study showed the level of urea in table (4-4) in control group was (53.15±1.052 mg/dl) while after treated with lead was (30.00±1.309 mg/dl) which didn't have significant increased compared with control group. This result of urea after exposure to lead were disagreement with (Mohammed *et al*., 2017) which noted the level of urea and creatinine increased in the serum after lead administration in rats.

The level of urea after treated with *Nerium oleander* was (77.50±13.098 mg/dl) which have significant increased compared with control group. This result may be due to damage in kidney or inflammation in kidney tissue lead to enzyme leak to blood circulation. This physiologic change disagreement with (Radostits and Gay, 2007) which noted increased the level of serum urea after treated with *Nerium oleander*.

The level of urea after treated with compound have significant increased (87.20±12.578 mg/dl). The value of urea after treated with (lead+ *Nerium oleander*) have prominent effects on kidney lead to increased level of urea in blood.

4.2.4 The Biochemical Results of Creatinine

Table (4-4) showed the normal value of creatinine in the serum of white mice in control group (0.417±0.0046 mg/dl) . The level of creatinine after treated with lead was (0.297±0.0090 mg/dl) which have significant decreased compared with control group, this result were disagreement with (Lee *et al*., 2014) which observed the accumulation of lead in kidney tissue caused damage and dysfunction of kidney lead to increased level of creatinine in serum.

The creatinine level after treated with *Nerium oleander* was (0.569±0.0427 mg/dl) which have significant increased compared with control group. This biochemical change may because of acute damage in glomeruli and renal tubules lead to increase in creatinine excretion in kidney. This result was constant with (Rahymah *et al*., 2011) which showed the value of creatinine increase after exposure to *Nerium oleander* toxicity.

The level of creatinine after oral administration with compound lead and compound was (0.574±0.0410 mg/dl) have significant increased with control group.

4.2.5 The Results of Hematological test

A- The Results of W.B.Cs

In table (4-5) showed a count of W.B.Cs in control group was (2.015±0.087). The total counts of W.B.Cs was (3.876±0.306) after treated with lead which have significant increased compared with control group. This result agreement with (Guidotti *et al*., 2008) which noted the count W.B.Cs after oral administration with lead was significant increased in blood of male albino rats.

The total of leukocyte after exposure to *Nerium oleander* was (3.840±0.097) which have significant increased compared with control group. This physiologic change may be due to inflammation in organs, injury in W.B.Cs and damage in bone marrow lead to this change. This result confirmed with (Radostits and Gay, 2007) which showed the leukocyte increased after treated with *Nerium oleander* aqueous leaf extract in Rabbits.

The count of W.B.Cs after treated group with compound lead and *Nerium oleander* was (5.113±0.252) that have significant increased compared with control group, this mix of (lead and *Nerium oleander*) have high effect on the level of leukocyte in blood.

B- The Results of R.B.Cs

Table (4-5) showed the count of erythrocyte in control group was (7.130±0.385). R.B.Cs count after exposure to lead was (6.334±0.220) which have significant decreased compared with control group. This change in count of R.B.Cs may be due to acute hemorrhage and iron deficiency lead to this result. This biochemical result similar to (Ibrahim *et al*., 2012) which noted treated rats with lead caused reduction in Hb concentration and W.B.Cs count. The count of R.B.Cs after treated with *Nerium oleander* was (6.591±0.107) which didn't have significant differences between control and *Nerium oleander* group. This result in count of R.B.CS disagreement with (Abdelouahab *et al*., 2011) which showed decrease in H.G.B, R.B.C and P.V.C after alkaloid extracts of seeds of Datura stramonium on mice.

The value of erythrocyte after treated with lead and *Nerium oleander* was (6.779±0.129) which didn't have significant differences compared with control group.

C- The Results of H.G.B

The result in table (4-5) showed the concentration of H.G.B in control group was (11.869±0.403g/dl). The biochemical result of H.G.B in white mice after treated with lead was (8.265±0.447g/dl) which have significant decreased compared with control group. This result may be due severe hemorrhage or lead caused disrupt production of R.B.Cs lead to reduction in H.G.B in blood. This result confirmed with (Azoz and Raafat, 2012) which noted exposure of blood to lead causes a decrease in hemoglobin value .

The biochemical result of H.G.B after treated with aqueous extract of *Nerium oleander* was (9.609±0.191g/dl) that have significant decrease compared with control group. This change in hemoglobin value agreement with (Adam *et al*., 2002) which showed The values of Hb and PCV were significant decrease.

The result of H.G.B after treated group with compound was (10.354±0.208g/dl) which have significant decreased compared with control group.

D- The Results of P.L.T

The physiologic result of P.L.T count in control group was (888.03±21.570). The value of P.L.T after exposure to lead was (665.12±84.441) which have significant decreased compared with control group. This change in result may be due to iron deficiency which enters into synthesis of hemoglobin, defect in bone marrow and hemorrhage lead to destruction in platelets. This result constant with (Vutukuru, 2005) which noted the count of P.L.T reduction in Indian carp after treated with Hexavalent Chromium.

The count of platelets after treated group with *Nerium oleander* was (847.58±65.067) which didn't have significant differences between control and *Nerium oleander*. This result non confirmed with (Al-Naqqash *et al*., 2013) which noted significant increase in MCV, P.L.T after treated with *Nerium oleander* Ethanolic Extract in Mice.

Platelets count after treated with compound lead and *Nerium oleander* was (880.08±17.486) that didn't have significant differences between treated group with compound and control.

E- The Results of L.Y.M

Table (4-5) that appeared the value of lymphocyte a count, the lymphocyte a count in control group was (2.318±0.061), while the lymphocyte count after treated with lead was (3.154±0.169) which have significant increased compared with control group. This result may be due to inflammation in blood or organs and injury in spleen and liver. This result agreement with (Lee *et al*., 2019) which showed organ destruction in fish exposed to lead caused lead to an increase in L.Y.M.

Physiologic change in value of lymphocyte after treated with *Nerium oleander* was (1.912±0.088) which didn't have significant differences in both control group and treated group with *Nerium oleander*. This result of count L.Y.M dis agreement with (Akhtar *et al*., 2014) which showed elevation count of lymphocyte after exposure to *Nerium oleander* extract in wistar rats.

The lymphocyte a count after treated with lead and watery extract of *Nerium oleander* which lead to significant increased in the lymphocyte a count (5.166±0.222), this result have significant increased compared with control group.

Table (4.4) : Level of biochemical parameters in serum of treated group M± S.D

Treatment	AST U/L	ALT U/L	Urea mg/dl	Creatinine mg/dl
control	365.15±12.295^{b}	42.00±1.891^{d}	53.15±1.052^{bc}	0.417±0.0046^{b}
Lead	609.20±14.746^{a}	143.35±10.957^{b}	30.00±1.309^{c}	0.297±0.0090^{c}
Nerium oleander	481.55±26.238^{ab}	84.70±7.142^{c}	77.50±13.098^{ab}	0.569±0.0427^{a}
Compound	485.45±80.927^{ab}	216.20±20.666^{a}	87.20±12.578^{a}	0.574±0.0410^{a}

Different letters vertically indicate the existence of significant differences between the averages at the possibility of (0.01)

(Table4.4-1) Results of ANOVA analysis of AST enzymes in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
AST	Between Groups	596026.238	3	198675.413	5.224	.002
	Within groups	2890435.650	76	38032.048		
	Total	3486461.888	79			

Duncan

TRT	N	Subset for alpha = 0.01	
		1	2
Control	20	365.1500	
Nerium oleander	20	481.5500	481.5500
Compound	20	485.4500	485.4500
Lead	20		609.2000
Sig.		.068	.053

(Table4.4-2) Results of ANOVA analysis of ALT enzymes in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
ALT	Between Groups	342399.737	3	114133.246	37.932	.000
	Within groups	228673.950	76	3008.868		
	Total	571073.688	79			

Duncan

TRT	N	Subset for alpha = 0.01			
		1	2	3	4
Control	20	42.0000			
Nerium oleander	20		84.7000		
Lead	20			143.3500	
Compound	20				216.2000
Sig.		1.000	1.000	1.000	1.000

(Table4.4-3) Results of ANOVA analysis of urea in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
Urea	Between Groups	39552.138	3	13184.046	7.927	.000
	Within groups	126396.750	76	1663.115		
	Total	165948.887	79			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Lead	20	30.0000		
Control	20	53.1500	53.1500	
Nerium oleander	20		77.5000	77.5000
Compound	20			87.2000
Sig.		.077	.063	.454

(Table4.4-4) Results of ANOVA analysis of creatinine enzymes in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
Creatinine	Between Groups	1.061	3	.354	19.578	.000
	Within groups	1.373	76	.018		
	Total	2.434	79			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Lead	20	.2975		
Control	20		.4170	
Nerium oleander	20			.5690
Compound	20			.5740
Sig.		1.000	1.000	.907

Table (4.5) : Level of hematological parameters of treated group M± S.D

Treatment	Control	Lead	*Nerium oleander*	Compound
W.B.C 10e3/uL	2.015±0.087^{c}	3.876±0.306^{b}	3.840±0.097^{b}	5.113±0.252^{a}
R.B.C 10e6/uL	7.130±0.385^{a}	6.334±0.220^{b}	6.591±0.107^{ab}	6.779±0.129^{ab}
H.G.B g/dL	11.869±0.403^{a}	8.265±0.447^{c}	9.609±0.191^{b}	10.354±0.208^{b}
P.L.T 10e3/uL	888.03±21.570^{a}	665.12±84.441^{b}	847.58±65.067^{a}	880.08±17.486^{a}
L.Y.M %	2.318±0.061^{c}	3.154±0.169^{b}	1.912±0.088^{c}	5.166±0.222^{a}

Different letters vertically indicate the existence of significant differences between the averages at the possibility of (0.01).

(Table4.5-1) Results of ANOVA analysis of W.B.C in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
W.B.C	Between Groups	97.752	3	32.584	69.051	.000
	Within groups	35.863	76	.472		
	Total	133.615	79			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Control	20	2.0150		
Nerium oleander	20		3.8405	
Lead	20		3.8765	
Compound	20			5.1135
Sig.		1.000	.869	1.000

(Table4.5-2) Results of ANOVA analysis of R.B.C in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
R.B.C	Between Groups	6.734	3	2.245	1.992	.122
	Within groups	85.631	76	1.127		
	Total	92.364	79			

Duncan

TRT	N	Subset for alpha = 0.01	
		1	2
Lead	20	6.3340	
Nerium oleander	20	6.5910	6.5910
Compound	20	6.7790	6.7790
Control	20		7.1300
Sig.		.216	.134

(Table4.5-3) Results of ANOVA analysis of H.G.B in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
H.G.B	Between Groups	135.556	3	45.185	20.393	.000
	Within groups	168.399	76	2.216		
	Total	303.955	79			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Lead	20	8.2655		
Nerium oleander	20		9.6090	
Compound	20		10.3545	
Control	20			11.8690
Sig.		1.000	.117	1.000

(Table4.5-4) Results of ANOVA analysis of P.L.T in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
P.L.T	Between Groups	659703.017	3	219901.006	3.624	.017
	Within groups	4611418.475	76	60676.559		
	Total	5271121.492	79			

Duncan

TRT	N	Subset for alpha = 0.01	
		1	2
Lead	20	665.1265	
Nerium oleander	20		847.5840
Compound	20		880.0840
Control	20		888.0335
Sig.		1.000	.629

(Table4.5-5) Results of ANOVA analysis of L.Y.M in study groups of mice

ANOVA						
		Sum of squares	df	Mean square	F	Sig.
L.Y.M	Between Groups	125.770	3	41.923	93.234	.000
	Within groups	34.174	76	.450		
	Total	159.944	79			

Duncan

TRT	N	Subset for alpha = 0.01		
		1	2	3
Nerium oleander	20	1.9125		
Control	20	2.3185		
Lead	20		3.1540	
Compound	20			5.1665
Sig.		.059	1.000	1.000

5.1 Conclusions

1- The effect of *Nerium oleander* on the brain lead to cells hypertrophy and Apoptosis in the different regions of gray matter in the cerebrum of brain. The effect of *Nerium oleander* on the small intestine, which lead to exfoliated the epithelial layer that lined the internal intestinal lumen and tunica muscularis was spirated into three thin layers, while the effect of *Nerium oleander* on the liver caused acute cellular destruction and degenerative in portal area, also effect of *Nerium oleander* on the kidney, which lead to acute degeneration in the wall of P.C.T and the glomerular capillaries have prominent lumen. The results explain the effect of *Nerium oleander* on biochemical results showed significant increase in the level of ALT and creatinine while the effect of *Nerium oleander* on the hematological parameters noted significant decrease in the count of H.G.B and significant increase in the count of W.B.C.

2- The lead has an impact role on the brain, which lead to de vacuoles in many location of brain and blood hemorrhage of brain tissue while the effect of lead on the intestine caused destruction in epithelial layer and the core of villi have prominent tissue degeneration. The effect of lead solution on the liver, which caused blood hemorrhages area in liver parenchyma and wide empty cystic dilation while the effect of lead on the kidney, which lead to the Bowman's space in the most renal corpuscle filled with thick secretion and noted abnormal Henle loop branches. The effect of lead on the biochemical result showed significant increase in the level AST, ALT and significant decrease of creatinine, also noted the effect of lead on hematological parameters which showed significant decrease in count of R.B.Cs, H.G.B and P.L.T and significant increase in count of L.Y.M and W.B.C .

3- Mixed of (lead+*Nerium oleander*) have real effects on the brain showed hemolysis spots and high cellular proliferation of inflammatory cells while the effect of compound lead and *Nerium oleander* on the small intestine, which lead to completely the separated the submucosal layer from tunica muscularis and to modification in the shape of epithelial cells. The effect of lead and *Nerium oleander*

solution on the liver caused abnormal portal area filled with blood and hepatocyte loss their nuclei, also effect of compound on the kidney, which lead to renal corpuscles have prominent destruction in the glomerular capillaries and acute destruction in the epithelial layer. Also the effect of compound solution on the physiologic changes showed significant increase in the level of ALT, urea and creatinine while the effect of the compound on the count of hematological parameters showed significant increase in the level of W.B.C and L.Y.M and significant decrease in the count of H.G.B.

Recommendations

1-Activating laws to prevent and limit using lead & *Nerium oleander* in treatment and spread information about its impact.

2- Currying out studies about effects of lead and *Nerium oleander* in region that uses it.

3- Reducing spreading of *Nerium oleander* and the media raising awareness about the dangers and toxicity of this plant.

4- Conducting ultra-histological study of the effects of lead and *Nerium oleander*.

5- Conducting Immunohistochemical studies of lead in different organs.

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الخلاصة

تهدف الدراسة الحالية إلى التركيز على التغيرات النسيجية والكيموحياتية في بعض الاعضاء للفئران البيضاء بعد معاملتها بالرصاص ونبات الدفلة وهو التركيب الرئيسي للسقوة.

حيوانات التجربة قسمت إلى أربعة مجموعات، ثلاث منها هي المجاميع المعالجة (B,C وD) كل منها تمت معاملتها بمركب مهم من السقوة، بينما المجموعة (A) تعتبر مجموعة السيطرة.

النتائج النسيجية الرئيسية تضمنت تجمع الخلايا الإلتهابية و إنحلال في نسيج الدماغ. المقاطع النسيجية للدماغ بعد معاملتها بالدفلة أظهرت تضخم في الخلايا العصبية، نزيف دموي، أحتقان دموي، وتنخر في في نسيج الدماغ، لذلك أظهرت موت مبرمج للخلايا في مناطق مختلفة من الدماغ، بينما، نسيج الدماغ بعد معاملته بالرصاص والدفلة لوحظ نقاط تحلل دموي، وتجمع خلوي للخلايا الإلتهابية.

التغيرات النسيجية في الأمعاء الدقيقة بعد معاملتها بالرصاص أظهرت دمار بارز في طبقة الغشاء المخاطي والخلايا الطلائية فقدت أنويتها، ظهر في تجويف الأمعاء بعد معاملته بالدفلة إنفصال في الطبقة الطلائية و العضلية فصلت إلى ثلاث طبقات نحيفة، بينما نتيجة الامعاء بعد معاملتها بالرصاص والدفلة أظهرت إنفصال تام للطبقة تحت المخاطية من طبقة الغلالة العضلية وتحطم الزغابات.

النتائج النسيجية للكبد بعد معاملته بالرصاص والدفلة لوحظ نزيف في النسيج الحشوي، الكبد فيه فراغات واسعة تحتوي كريات دم حمراء على التوالي. الكبد بعد معاملته بمركب الرصاص والدفلة ظهر المنطقة البوابية غير طبيعية ومملوءة بالدم.

نتيجة الكلية بعد معاملتها بالرصاص أظهرت تحطم في الشعيرات الدموية الكبيبية ونزيف دموي، المقطع النسيجي بعد معاملته بالرصاص وبالرصاص والدفلة اظهر تحطم في منطقة القشرة، وإنحلال في جدار (P.C. T. و D. C. T.) وإنتشار خلوي غير طبيعي في القشرة والكريات الكلوية فيها تحطم بارز في الشعيرات الدموية الكبيبية على التوالي.

النتائج الكيموحيوية لـ (AST, ALT, urea, creatinine, C.B. P.) بعد معاملة المجاميع الثلاث بـ (الدفلة والرصاص وبالرصاص والدفلة) أظهرت فرق معنوي مقارنة بمجموعة السيطرة.

جمهورية العراق
وزارة التعليم العالي والبحث العلمي
جامعة المثنى / كلية العلوم
قسم علوم الحياة

دراسة نسجية ودمية وكيمو حيوية لبعض مكونات السقوة التي تؤثر على بعض الاعضاء في ذكور الفئران البيضاء

الرسالة مقدمة كجزء من متطلبات نيل درجة الماجستير في

علوم الحياة

من قبل

بشار عبد الله عبد الهادي

بكالوريوس علوم حياة /2008

بإشراف

أ. د. باسم عبد الله جاسم

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