



***Research Paper***

***Aloe vera* PROTECTS THE ALUMINIUM INDUCED DEGENERATIVE CHANGES IN LIVER AND KIDNEY OF ALBINO RATS, *Rattus norvegicus***

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**Abstract**

The current study was carried out to investigate the protective role of *Aloe vera* plant extract on aluminium induced degenerative changes in liver and kidney of albino rats. *Aloe vera* is a medicinal plant belonging to the family *-Liliaceae*, which has a wide range of therapeutic applications such as wound healing, diabetes, burns, for easing intestinal, curing ulcers and arthritic swellings. 30 adult rats were taken and divided into 3 groups 10 (5+5) for each. Control group animals were fed with normal diet and water *adlibitum*, as Group I or control group. Group II animals were fed with normal diet and received aluminium in a dose of 98 mg/kg of body weight orally for 30 and 60 days. Group III were fed with normal diet and received aloin (100mg/kg body weight) and aluminium sulphate (98 mg/kg body weight) for 30 and 60 days. On the last day of the experiment animals were sacrificed by cervical dislocation on 30<sup>th</sup> and 60<sup>th</sup> days respectively. While examination of liver and kidney sections, aluminium treated group *per se* showed abnormal characteristic architecture compared with the control group. Animals treated with aluminium along with the aloin showed recovery of liver tissue structure and kidney, which was more prominent in long time treatment in comparison to the short time treated group. It is also interesting to note that protective effect of aloin against aluminium toxicity was more effective in the 60 days of treatment as compared to the 30 days of aloin treatment.

Key words: Aluminium Toxicity, Aloe Vera, Albino Rats, Liver, Kidney and Histopathology.

**INTRODUCTION**

Aluminium, the third most abundant element of the Earth's crust, is a non-essential and toxic metal in humans [1]. Aluminium compounds have a wide range of uses in industrial, domestic, consumer, and medicinal products. Aluminium enters the human body via food, air, water and drugs<sup>1</sup>, and is present in many manufactured foods such as processed cheese, baking powders, cake mixes, frozen dough, pancake mixes [2] and pharmaceutical products, especially antacids[3]. In recent years, various epidemiological studies have suggested that Al plays a pathogenic role in Alzheimer's Disease (AD) [4]. Aluminium has a negative impact on human health[12]. Due to its abundance, every organism contains small quantities of aluminium and it can be found in practically all of the tissues of mammals, including the brain, liver, kidney, heart, blood, and bones [7]. Aluminium accumulation in the liver leads to cholestasis[5]. Chronic exposure to aluminium is involved in neuro-degenerative disorders, such as Alzheimer's

disease[17] dialysis, Parkinson's dementia,[8] and hepatotoxicity [9]. The toxic effects of aluminium appear to be mediated, at least in part, by free-radical generation [10]. The treatment commonly used in aluminium disorders is desferrioxamine[11] However, desferrioxamine therapy is associated with undesirable side effects, it is very expensive, and it is only efficient when applied intravenously or subcutaneously[12].

Plants have been used to treat various diseases and have been an exemplary source of medicine over the years [13]. It has been reported that plant extracts detoxify various kinds of environmental pollutant [14]. *Aloe vera* is one such ancient plant whose medicinal properties have been known since centuries and has wide range of therapeutic applications such as wound healing effect, reduction of blood sugar in diabetes, for soothing burns, for easing intestinal, for curing ulcers and for reducing arthritic swellings [[15, 16]. *A. vera* gel contains anthroquinones (aloin, aloe-emodin) which may have a variety of properties of anti oxidant agent, including the protective role for heavy metal toxicity [17] The goal of this study was to investigate the protective role of *Aloe vera* on aluminium induced degenerative changes in liver and kidney of rats.

## MATERIALS AND METHODS

Healthy adult albino rats (*Rattus norvegicus*) weighing  $175 \pm 5$  gm were used for the experiments, procured from Mhow, Bhopal (MP) India. And maintained in our laboratory. The rats were acclimatized in laboratory conditions for two weeks and were maintained at  $28 \pm 2^{\circ}\text{C}$  room temperature and relative humidity ( $60 \pm 10\%$ ) with a 12 hours light-dark cycle in the animal house of biotechnology laboratory, Saifia Science College, Bhopal. Food and water were provided *ad libitum* throughout the experiment to avoid effects of starvation. No mortality was observed during the acclimatization period and during whole experimentation period up to 60 days.

### Collection and preparation of plant materials for experiment

*Aloe vera* plant leaves were used for the present study. Leaves of *A. vera* were collected in and around the Bhopal. Preparation of *A. vera* (leaf gel) extracts was done according to the method of Arunkumar and Muthuselvam [20] with slight modifications. Skin of the leaves were peeled and the gel inside was used for extraction. 100 gms of the gel was added to 250 mL of ethanol and extracted using the Soxhlet assembly. Later on, the solvent of the extracted material was removed at low temperature in a rotary vacuum evaporator and the resulting dried extract was lyophilized in a freeze dryer.

### Experimental design

All the experimental animals were divided into three groups as group I,II and III.

**Group I:** - This group of 10 (5+5) animals was fed with normal diet and water *ad libitum*, as control group.**Group II:** - This group of 10 (5+5) animals was fed with normal diet and aluminium in a dose of 98 mg/kg of body weight orally for 30 and 60 days. **Group III:** - This group of 10 (5+5) animals were fed with normal diet and received aloin (100mg/kg body weight) and aluminium sulphate (98 mg/kg body weight) for 30 and 60 days. Animals were sacrificed by cervical dislocation on 30<sup>th</sup> and 60<sup>th</sup> days respectively. Liver and kidney were isolated and kept in ice cold conditions for experiment.

### Histopathological work

Samples of liver and kidney of rats both control and treated with aluminium sulphate *per se* treated and aluminium sulphate plus aloin treated rats were immediately excised, fixed in Bouins fixative followed by standard procedures of paraffin embedding and were sectioned at 5-6 microns. Sections were stained with haematoxylin and eosin and mounted with Canada balsam/DPX abd were examined by Olympus light microscope to evaluate histological changes. The experimental chemicals were obtained from Sigma Chemical Co. USA of analytical grade.

## RESULTS

In the present investigation, histological analyses of liver and kidney were done in albino rats subjected to different durations of aluminium sulphate administration. Photomicrographs were taken for histopathology of the liver and kidney tissue sections at 10X and 40X magnification and reported here.

### Liver

Histological examination of liver of control is showed normal morphology of hepatocytes, portal triads and also normal vasculature. The liver parenchyma of control rats showed a normal architecture composed of cords that extended from the central vein to the portal space. Hepatocytes were arranged in cords separated by capillary sinusoids lined with endothelial cells. Kupffer cells were also present along the sinusoidal capillary. Each hepatocyte is a polygonal cell with a large centrally located spheroid nucleus having a chromatin structure and a distinct nucleolus. The cytoplasm of hepatocytes is faintly granular.

While examination of liver sections of aluminium sulphate treated rats shows abnormal characteristic architecture compared with the control group. Histopathological examination of aluminium treated rats revealed remarkable degenerative changes such as diffuse disorganization of hepatic cords, vascular congestion, dilatation of sinusoids capillary, central vein and portal space. Mild lymphoid and mononuclear infiltrations were observed within the portal areas and central vein the wall of which was dislocated. Nuclei chromatin was fragmented and cytoplasm contained many vacuoles. Variations in the size of hepatocytes were also observed where some have deeply stained nuclei and homogeneous and acidophil cytoplasm. Cytoplasm of the hepatocytes are characterized by coarse, pink, darkly stained granules and few vacuoles in 30 days treated animals, while there is increase in cytoplasmic vacuolization in 60days treated animals. Inflammatory cellular infiltration was abundant around central vein which is more intense in liver sections of 60 days treated animals.

The photographic plate shows the nuclei of hepatocytes in aluminium sulphate treated rat *Rattus norvegicus* appeared larger and more irregular in shape with very little peripheral condensed chromatin. Clumped chromatin was also observed in many of the hepatocytes and some nuclei appeared pycnotic. The nucleoplasm of many cells showed continuity with the cytoplasm due to loss of nuclear membranes.

Histopathological examination of liver of aluminium with aloin treated rats showed hepatocytes, more or less similar to control group, with marked improvement in its histological structure in comparison to the Al *per se* treated group, represented by nil to moderate degree in degeneration of hepatocytes, fatty change in hepatocytes, coagulative necrosis in hepatocytes, diffused Kupffer cells proliferation in portal area, diffuse Kupffer cells proliferation in between hepatocytes, and dilatation in central vein.

### Kidney

The kidney tissue of aluminium sulphate treated rats showed some pathological changes when compared to control animals. In 30 days intoxicated rats, the kidney has shown reduced lumen of the proximal tubules. The changes were more intensified in 60 days multiple dose animals besides atrophy of glomeruli. Degeneration in proximal tubules, reduction in proximal tubule lumen, atrophy of glomeruli and necrotic changes in epithelial layer of proximal tubules was more prominent in 60 days treated animals with comparison to control animals and also 30 days treated one.

The kidney of treated group rats also showed some other significant histopathological abnormalities like disorganized glomeruli with distorted Bowman's capsule, loss of cellular integrity of tubular cells, multifocal cloudy and cellular infiltrations. The tubular cells appeared swollen with cytoplasmic vacuolization and coarse pink cytoplasmic granules especially in the

cells of the proximal and distal convoluted tubules. Lumens of these tubules were obliterated or highly reduced. It was found that supplementation of aloin with aluminium, protected the animals from toxic effects of the metal with normal appearance of glomeruli which did not show damage at the spots examined. Casts have been noticed to be absent and the tubules were compact, rounded and at places thin-walled but neither dilated nor damaged. It is also interesting to note that protective effect of aloin against aluminium toxicity was more effective in the 60 days of treatment as compared to the 30 days of aloin treatment.

## DISCUSSION

Previous studies on aluminium toxicity have been demonstrated that aluminium accumulation in liver is associated with a number of biochemical changes, which include the release of enzyme markers of liver injury, and alterations in the oxidant status [10]. Aluminium cause histopathological alterations in different tissues of albino mice. Our light microscopy findings following aluminium exposure of rat liver and tissues show structural changes in the sinusoids that are accompanied by hepatocyte column and cytoplasmic, nuclear, and cell membrane alterations in these hepatocytes, all of which support the previously reported biochemical alterations that have been reported to be due to aluminium damage to the liver. The treatment commonly used in aluminium disorders is desferrioxamine, which is a chelator that has a large capacity to decrease the aluminium body burden by increasing its excretion in the urine [11]. However, as indicated above, it is only efficient when it is applied intravenously or subcutaneously and it has been shown that desferrioxamine therapy has side effects that are often not well tolerated. Thus, its application limits the success of this therapy, while it has also been seen to be an expensive treatment [12].

*Aloe vera* plant extract has a wide range of therapeutic applications [16]. *A. vera* gel contains anthroquinones (aloin, aloe-emodin) which may have a variety of properties of anti oxidant agent, including the protective role for heavy metal toxicity [17]. Previous studies have also shown that as an antioxidant, plant extracts may improve the pro-oxidant effects of aluminium. Similarly, [36] have reported the nephroprotective activity of *Tephrosia purpurea* against arsenic-induced toxicity in rats. There was also the presence of vacuolar changes along with necrosis, desquamation and distorted cellular architecture.

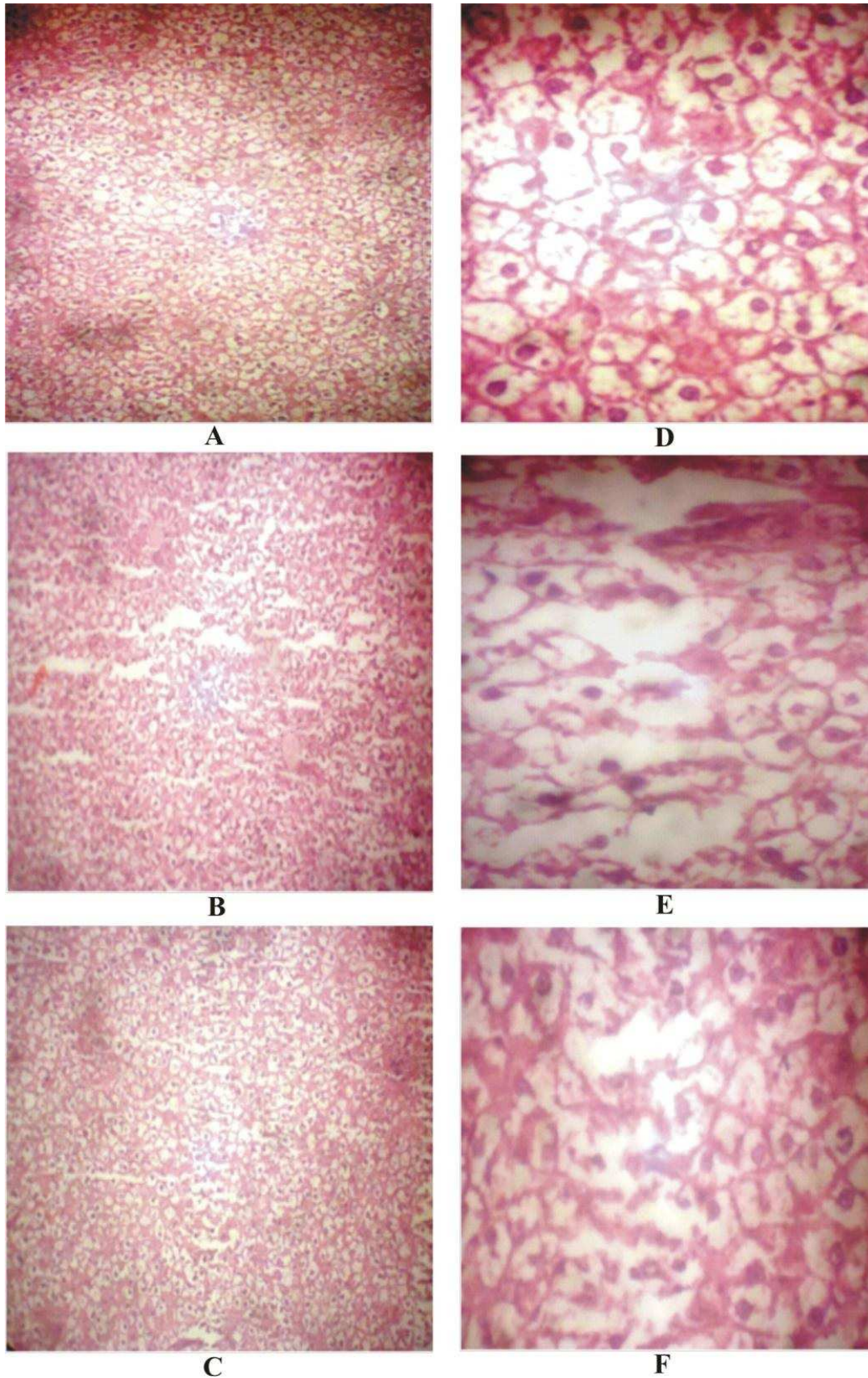
In the present study, *A. vera* plant extract of aloin was demonstrated to serve as an antioxidant, and to prevent the degenerative effects of aluminium on the microscopic morphology of rat liver and kidney tissues. The results of this study thus clearly demonstrate that Aloin co-treatment under aluminium exposure ameliorates the histological alterations in the liver and kidney tissue that is caused by aluminium exposure.

Recovery of liver tissue structure was more prominent in long time treatment of aloin in comparison to the short time treated group. In 60 days treated group, the liver tissues got restored most of their normal structure and were able to diminish the fibrosis, congestion, incidence of inflammatory cells infiltration, centrilobular hepatocytes swelling, hepatocyte vacuolization, fatty changes and hemorrhagic clots.

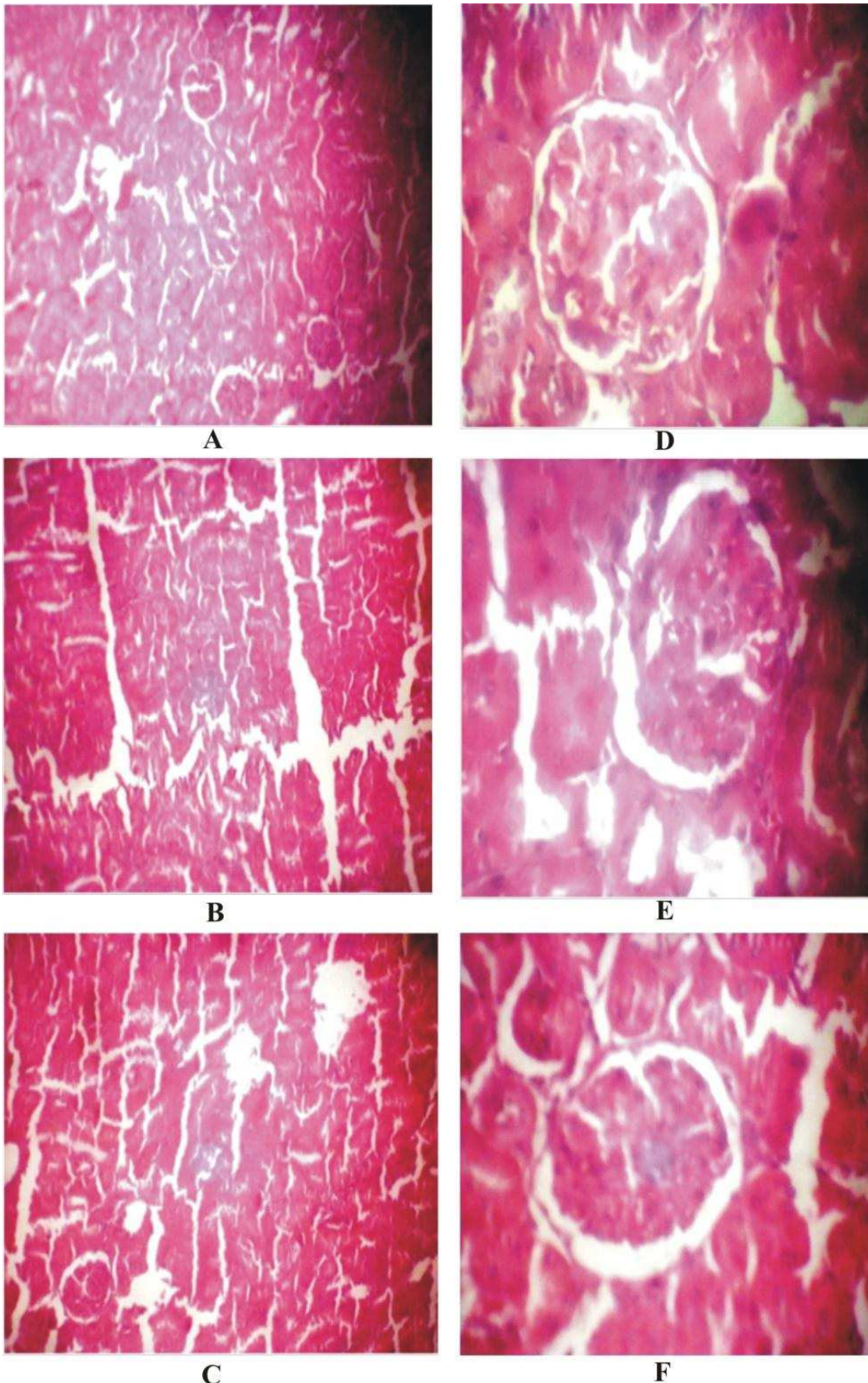
The kidney of rats treated with aluminium along with the aloin also showed lesser desquamation and necrotic changes in the 60 days of aloin and aluminium co-treated rats, indicating that aloin has a significant role in protecting the aluminium exposed animals from aluminium induced nephrotoxicity. Thus, this study indicates that protective role of aloin treatment against the adverse effects of aluminium on liver and kidney of albino rats.

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**Photomicrograph plate 1:** Histopathology of the liver tissue showing **A.** Control, **B.** Aluminium Sulphate *Per se* treated, **C.** Aluminium Sulphate + Aloin co-treated sections at 10X magnification and **D.** Control, **E.** Aluminium Sulphate *Per se* treated, **F.** Aluminium Sulphate + Aloin co-treated sections at 40X magnification.



**Photomicrograph plate 2:** Histopathology of the kidney tissue showing **A.** Control, **B.** Aluminium Sulphate *Per se* treated, **C.** Aluminium Sulphate + Aloin co-treated sections at 10X magnification and **D.** Control, **E.** Aluminium Sulphate *Per se* treated, **F.** Aluminium Sulphate + Aloin co-treated sections at 40X magnification.

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