The present investigation was aimed to evaluate the prophylactic role of *Moringa oleifera* against the histological and biochemical changes induced by mercuric chloride or radiation alone or in combination, in the renal lesions of Swiss albino mice.

Adult Swiss albino mice (6-8 weeks old) were procured from Lala Laljpat Rai University of Veterinary and Animal Sciences, Hisar. The animals were kept in polypropylene cages. They were fed with standard mice feed and water was given *ad libitum*. The cages were cleaned daily. The temperature of the room was maintained between 22-27°C.

The dried powder of *Moringa oleifera* was procured from the Umalaxmi organics private limited, Jodhpur (Raj) and aqueous extract of the same was obtained in the department. The plant extract of *Moringa* was fed orally at the dose of 150 mg/kg body weight. The *Moringa* extract was given daily from seven days prior to individual or combined treatment of mercuric chloride and radiation and continued up to the last autopsy interval.

In the recent years, immense interest has been developed in the field of chemoprotection against radiation and heavy metals induced changes. Therefore the present study was planned to investigate the protective effect of *Moringa oleifera* (a herbal drug) against combined exposure on kidney of Swiss albino mice.
PLAN OF WORK

In the present investigation, healthy adult male Swiss albino mice (6-8 weeks old) were used. In order to investigate the protective effect of *Moringa oleifera* the animals were divided into following groups:

Group – I  (Sham-irradiated animals) The animals of this group were sham- irradiated and served as control (normal) group.

Group - II  (Mercuric chloride treated animals)

All the animals of this group were orally fed with mercuric chloride solution at the dose of 0.5 ppm *ad libitum* in drinking water continuously till the end of experiment.

Group - III  (Irradiated animals)

The animals of this group were exposed to sub lethal doses of gamma radiation from Cobalt-60 source. This group was further divided into two sub-groups on the basis of radiation dose received:

Sub-group III a : 2.5 Gy
Sub-group III b : 5.0 Gy

Group - IV  (Animals treated with radiation and Mercuric chloride)

All the animals of this group were orally fed with mercuric chloride solution (0.5 ppm) and also exposed to different doses of gamma radiation. This group was further divided into two sub-groups on the basis of radiation dose received:
Group - IV (Sub-group IV a : 2.5 Gy + Mercuric chloride  
Sub-group IV b : 5.0 Gy + Mercuric chloride)

Group - V (Mercuric chloride and Drug treated animals)

The animals of this group were orally fed with Mercuric chloride (0.5 ppm) and also received *Moringa oleifera* orally for seven days at a dose of 150 mg/kg. body weight/animal/day prior to mercuric chloride treatment and continued up to the last autopsy interval.

Group - VI (Radiation and drug treated animals)

The animals of this group were exposed to gamma radiation from Co$^{60}$ source. The *Moringa oleifera* was given seven days prior to irradiation and continued up to last autopsy interval. This group was further divided into two sub-groups on the basis of radiation dose received :

Sub-group VI a : 2.5 Gy + *Moringa oleifera* 
Sub-group VI b : 5.0 Gy + *Moringa oleifera*

Group - VII (Radiation, Mercuric chloride and drug treated animals)

The animals of this group were orally fed with Mercuric chloride solution at the dose rate of 0.5 ppm and received *Moringa oleifera* orally (150 mg/kg. body weight/animal/day) for seven days prior to irradiation and mercuric chloride till the last autopsy day of experiment.
This group was further divided into two sub-groups on the basis of radiation dose received:

Sub-group VII a : 2.5 Gy + Mercuric chloride + *Moringa oleifera*

Sub-group VII b : 5.0 Gy + Mercuric chloride + *Moringa oleifera*

A minimum of five animals from groups II to VII sacrificed by cervical dislocation and autopsied and each post treatment intervals 1, 2, 4, 7, 14 and 28 days. The weight of animals was recorded before the autopsy. The following studies were taken into consideration:

1. **Reno-somatic index**

   The weight of kidney shall be recorded and expressed as Reno-somatic index.

   
   \[
   \text{Reno-somatic index} = \frac{\text{Weight of kidney}}{\text{Weight of body}} \times \frac{100}{1}
   \]

   (gm/100gm of body wt.)

2. **Histological studies**

   After sacrificing the animals, pieces of the kidneys were taken out and some of them were immediately fixed for histological observations by routine procedure.
3. Biochemical studies

The following biochemical parameters were taken into consideration:

1. Total proteins [Lowry et al., 1951]
2. Glycogen [Montogomery, 1957]
4. Acid phosphatase [Fiske and Subbarow, 1925]
5. Alkaline phosphatase [Fiske and Subbarow, 1925]
6. DNA [Ceriotti, 1952]
7. RNA [Ceriotti, 1955]

OBSERVATION

Reno somatic index

In the present investigation, the value of reno-somatic index was increased in all the groups. In the groups II, III and IV the value of reno-somatic index increased up to day-14 thereafter it declined on day-28. Whereas in the groups V, VI and VII the value of reno-somatic index rose up to day-7 thereafter it declined on day-14 and continued to decline up to day-28. The combined treatment of radiation and Mercuric showed synergistic effect. In Moringa oleifera treated groups there was a lesser increase in these value showing protective effect of Moringa oleifera. These observation coincided with histopathological and biochemical observation.
Histopathological

In the present experiments histopathological changes were found dose dependent in the kidney of Swiss albino mice exposed to 2.5 or 5.0 Gy gamma rays with or without Mercuric chloride treatment. The changes observed on day-1 after exposure to 2.5 Gy were distortion of renal architecture, intracellular oedema, hyperplasia, cytoplasmic degranulation, vacuolation and pycnotic nuclei blurred renal tubules and damaged glomeruli. The changes were more marked on day-4 and continued up to day-14. But on day-28 the signs of recovery were observed. After exposure to a higher dose (5.0 Gy) similar changes were noticed but they were more pronounced and there was late manifestation of recovery. In the combined treatment of radiation and mercuric chloride synergistic effects were observed. The kidney of Moringa oleifera treated animals exhibited less severe damage as compared to non-drug treated animals at all the corresponding intervals. An early and fast recovery was also noticed in Moringa oleifera pretreated animals.

BIOCHEMICAL

Total Proteins

In the present study, the total protein content of the mouse kidney showed an increasing trend in non-drug treated groups III and IV as well as Moringa oleifera treated groups VI and VII respectively. In the groups II and V the value of total protein showed a decreasing trend. This increase or decrease was comparatively lesser in the Moringa oleifera treated animals showing protection by Moringa oleifera. The
value of total proteins increased in groups III and IV on day-1 which continued up to day 14 thereafter it declined on day-28, whereas, in groups VI and VII the value increased up to day-7 thereafter it declined on day-14 which continued up to day-28. Similarly in group II the value decreased up to day-14 thereafter it increased on day-28, whereas in the group V the value decreased up to day-7 thereafter it increased on day-14 which continued up to day-28. The combined action of Mercuric chloride and radiation showed synergistic effect.

**Glycogen**

The value of glycogen showed an increasing trend in all the non-drug treated groups as well as *Moringa oleifera* treated groups. This increase was comparatively lesser in the *Moringa oleifera* treated animals showing protection by *Moringa oleifera*. The value of glycogen increased in groups II, III and IV on day-1 which continued up to day-14 thereafter it declined on day-28, whereas in *Moringa oleifera* treated groups V, VI and VII the value increased up to day-7 thereafter it declined on day-14 which continued up to day 28 without reaching to the normal. The combined action of Mercuric chloride and radiation showed synergistic effect.

**Cholesterol**

The value of cholesterol decreased on day-1 which continued up to day-14 in groups II, III and IV respectively. Thereafter it increased on day-28 without reaching to the normal level. In the *Moringa oleifera* treated groups V, VI and VII the value decreased up to day-7, thereafter it increased on day-14 and continued so up to day-28. The decrease was
found dose dependent. In *Moringa oleifera* treated groups a less severe decrease and early recovery in the cholesterol level was observed.

**Acid Phosphatase Activity**

The acid phosphatase activity increased on day-1 which continued up to day-14 in the groups II, III and IV respectively. The value decreased on day-28. In the *Moringa oleifera* treated groups V, VI and VII the value of acid phosphatase activity increased up to day-7 and then decreased on day-14 which continued up to day-28, but still the difference in the value was significant as compared to the normal group.

**Alkaline Phosphatase Activity**

The alkaline phosphatase activity increased on day-1 in the groups II, III and IV which continued up to day-14, thereafter decline on day-28 without reaching to the normal. Similarly in the *Moringa oleifera* treated groups V, VI and VII the value increased up to day-7 thereafter it decreased on day-14 and continued to decrease up to day-28. A less severe increase was observed in the drug treated groups.

**DNA**

The DNA content decreased in the kidney of Swiss albino mice in all the groups. The decrease was found dose dependent. The value of DNA declined up to day-14 in groups II, III and IV thereafter, it increased on day-28. In the *Moringa oleifera* treated groups V, VI and VII the DNA content decreased up to day-7 then it increased on day-14
which continued up to day-28. In *Moringa oleifera* treated animals there was lesser decrease and early recovery observed.

**RNA**

The concentration of RNA increased in kidney of all the groups. The RNA content increased on day-1 and continued up to day-14 significantly (P<0.001) in the non drug treated groups II, III and IV respectively. The value declined on day-28 without reaching to the normal level. In the *Moringa oleifera* treated groups V, VI and VII the RNA content increased up to day-7 significantly (P<0.001) thereafter it declined on day-14 which continued up day-28. The difference in the value was significant (P<0.001) as compared to the normal. The increase was found dose dependent. In the *Moringa oleifera* treated groups less severe increase was observed showing protection by *Moringa oleifera*.

*From the present findings following could be deduced –*

1. The kidney of Swiss albino mice suffered with radiation and mercuric induced changes at histological and biochemical levels.

2. Increase in reno-somatic index during early intervals may be due to fatty degeneration, increase in protein and glycogen levels and certain other factors, which have not been studied, in the present case.

3. Alterations in the biochemical parameteres followed the histological changes.
4. The combined treatment of radiation and mercuric chloride showed synergistic changes.

5. The kidney of *Moringa oleifera* treated animals showed less severe radiolesions and early and fast recovery in comparison to non-drug treated animals. Thus, it seems that *Moringa oleifera* has protected the kidney at both the dose levels with and without mercuric chloride treatment.

6. The *Moringa oleifera* might have protected the animals from radiation by more than one mechanism due to multiplicity of its properties.

7. Thus, *Moringa oleifera* is a good herbal radioprotector and can be given to cancer patients during radiotherapy to minimize the side effects of exposure.