Radiation damage to cells in the body can happen after a person receives radiation therapy to treat cancer. It can also happen if a person is exposed to radiation through x-ray imaging, nuclear power, or fallout from nuclear weapons. If severe enough, radiation damage may cause cancer, birth defects, heart disease, and other serious health problems. Doctors try to protect people undergoing radiation therapy for cancer by using low doses, being precise about targeting the radiation, and minimizing side effects. Usually side effects go away after the treatment stops.

X-ray photons carry enough energy to ionize atoms and disrupt molecular bonds. This makes it a type of ionizing radiation, and therefore harmful to living tissue. A very high radiation dose over a short amount of time causes radiation sickness, while lower doses can give an increased risk of radiation-induced cancer. In medical imaging this increased cancer risk is generally greatly outweighed by the benefits of the examination. The ionizing capability of X-rays can be utilized in cancer treatment to kill malignant cells using radiation
therapy. It is also used for material characterization using X-ray spectroscopy.

Attenuation length of X-rays in water showing the oxygen absorption edge at 540 eV, the energy\(^{-3}\) dependence of photo absorption, as well as a leveling off at higher photon energies due to Compton scattering. The attenuation length is about four orders of magnitude longer for hard X-rays (right half) compared to soft X-rays (left half).

Hard X-rays can traverse relatively thick objects without being much absorbed or scattered. For this reason, X-rays are widely used to image the inside of visually opaque objects. The most often seen applications are in medical radiography and airport security scanners, but similar techniques are also important in industry (e.g. industrial radiography and industrial CT scanning) and research (e.g. small animal CT). The penetration depth varies with several orders of magnitude over the X-ray spectrum. This allows the photon energy to be adjusted for the application so as to give sufficient transmission through the object and at the same time good contrast in the image.

X-rays have much shorter wavelength than visible light, which makes it possible to probe structures much smaller than what can be seen using a normal microscope. This can be used in X-ray microscopy to acquire high resolution images, but also in X-ray crystallography to determine the positions of atoms in crystals.
Radiation effects on animal kidney

Radiation injury to the kidney is a very important field for investigation as kidney is adapted for filtering wastes from the blood. Proper knowledge of the response by the kidney to ionizing radiation appears as a problem of great clinical and biological importance. But perhaps in no field of radiation biology are opinions so greatly divergent as in considering the kidney to be radio resistant.

Berdjis (1960); Rosen et al. (1960, 1961, 1964); Consgrove et al., (1965); Crummy et al., (1965); Bajarnason and Warren (1968) have implied that glomerular lesions and vascular changes are the basic features of radiation damage.

One of the earliest reports was the presence of albuminuria noted by Bearman and Linser (1904) in rabbit and by Buschke and Schmidt (1905) in guinea pigs and rabbits. Schulz Hoffmann (1905) described the early changes in rabbit kidneys as principally those of haemorrhage, glomerular congestion, transudation of the interstitial tissue and less frequently, severe tubular damage.

Many works followed in which an attempt was made on the kidney of mice, rats, rabbits guinea pigs and dogs by Warthin (1907). He irradiated the kidneys through the abdominal wall for 1 hour or less. Distinct but temporary cytological changes were seen in the tubular epithelium, consisting of swelling, vacuolization and some nuclear modifications.
Gabriel (1926) found the primary changes in the vascular system with secondary tubular damage and glomerular atrophy. Hartman et al, (1927) and Hartman (1939) in a series of experiments studied the clinical and pathological changes induced by radiation in dog kidneys and made major contribution to the study of radiation induce kidney diseases in dogs. It was evaluated that in some cases kidney may represent a fairly radiosensitive organ (under 2000 r) while in others, animal, died within short time (3000 r) depending upon the animal, type of radiation and dose of radiation. Ionizing radiation produced in the kidney morphological as well as functional changes, which are partly the result of a direct action of irradiation on the renal parenchyma and partly indirectly produced by toxic substance formed in or released from kidney cells under the impact of irradiation. These experiments were carried by Impiobato (1935).

The damage reported by Lacassagne (1946) in mice consisted of atrophy and degeneration with destruction of a number of convoluted tubules but no significant glomerular changes were recorded.

Metcalf (1954) found no evidence of histological changes in chronically irradiated rats and rabbits for 2 years, either with 1.000 or 250 kv machines. All animals receiving 10 r per days died during the experiments. It was not known whether this had any effect on the kidneys.
Antopol and Glaubeck (1956) irradiated mice with 600 r total body irradiation and concluded that radiosensitivity changes with the age of the animal.

After whole body x-irradiation of mice, Berdjis et al. (1956, 1959) and Konh et al. (1957) observed that kidney changes also vary with the sex, greater weight loss.

Wilson et al. (1958) exposed both kidney of rat to 1100 r single dose and noted hypertension and nephrosclerosis for a period of 4 to 7 months. If one kidney was irradiated, the animal developed nephrosclerosis of the kidney and hypertension. The other kidney showed minimal scarring or no lesions. If both kidneys were irradiated in situ, the rats developed hypertension and nephrosclerosis. Thus irradiation elicits two distinct effects namely hypertension and sclerosis (fibrosis) which can occur concurrently or independently.

Studies on the kidney of different mammalian species after x-irradiation have demonstrated changes in the aspartic acid, glycine, leucine and alanine, this was demonstrated by Dragoni (1959).

Whereas, Teiger (1959) and Rich et al. (1961) did not find any histochemical change in the irradiated kidneys of rats. Decrease of alkaline phosphatase in kidney of rats was observed by Dragoni (1959), De Simone et al. (1960) & Rich et al. (1961).

Guttaman & Kohn (1960, 1963) and Guttaman et al. (1961) claimed that the progressive interstitial glomerulosclerosis is a natural
phenomenon occurring in old animals whether irradiated or not, but it is accelerated by irradiation. Cole & Rosen (1961) irradiated mice with LD$_{50}$ dose (850 r) and then removed one kidney 2, 4 and 9 week later.

(i) After 2 weeks of irradiation, a sharp decrease or absence of mitotic activity was seen in the group nephrectomized. However, there was no histological evidence of cell death in this group,

(ii) After 4 or 9 weeks of irradiation, when nephrectomy was done partial recovery was manifested by a number of abnormal mitotic figures.

It was observed that some of the functions of kidney like glomerular filtration rate (GFR) and renal flow (RF) increase with the increased dose of radiation. The experiment was carried by Zaruba (1965) in which he irradiated dogs with 400, 500, 600 and 800 r single dose total body exposure and examined kidney functions at various intervals of time from 1 to 42 days post interval, during first 4 days GFR and FR increased significantly with increasing dose.

The animal which had received 600 r dose developed acute radiation syndrome with increased filtration factor and progressive reduction of RF, while GFR variations were not significant. Later, Zaruba (1965) either irradiated both kidneys with doses ranging from 600 r to 2400 r or gave total body exposure to animals with the same doses and found no significant functional changes. However, in total
body irradiation he found a marked decrease in the activity of alkaline phosphatase in the renal cortex. Based on these finding Zaruba (1968) concluded that the functional changes are not due to the direct effect of radiation on renal tissue.

Alkaline Phosphatase variations have been studied by Pospisil & Zaruba (1963) in dogs irradiated with cobalt-60 (600 total body). They found an accumulation of alkaline phosphatase in the capillary endothelium of the medulla, but no alkaline phosphatase activity was found in the brush border of convoluted tubules, which is the normal site.

Work done with pigeons shows that birds are five time less sensitive to radiation than adult rats in them change in kidney and liver occurred to the same extent as that observed in intestine, spleen, bone marrow and sex organs, which is not true for mammals (Bacq and Alexander 1966). Nagai et al. (1954) irradiated the chick embryo with x-irradiation of 600 r and noted that pycnotic kidney cells were scattered all over the section.

Several alterations like increase in weights of heart, liver and kidney, reduction in protein content of these organ and decreased activity of leucine with concomitant increase of glutamic oxaloacetic transaminase (GOT) activity were observed in rats after total body 450 r irradiation and observed at day 2 post-irradiation.
Tortuosity of the interlobular arteries, narrowing and obstruction of some afferent arterioles, proliferation of the endothelial cells and sub-endothelial connective tissue, multiple scattered-hyalinized glomeruli with increased inter capillary PAS positive material, degeneration and disintegration of the tubular epithelial cells were among the major histological changes. Thompson and Mckey (1969) confirmed these observations.

Mostofi et al. (1964) studied the histological damage suffered by a kidney of dog after irradiation with 2,500 r. The early changes were found as moderate to severe congestion and various degrees of interstitial extravasations, fibroblast proliferation and flattening and atrophy of the epithelium along with granular and red cell casts at 4 weeks. But at 12 weeks to 72 weeks there could be seen marked degree of tubular atrophy, interstitial fibrosis and glomerular atrophy and hyalinization occasionally associated with fibrinoid degeneration of some glomarular tufts and small or larger blood vessels.

In another experiment Cosgrove et al. (1955, 1956) noted a high incidence of glomerulosclerosis in rodent, surviving more than one year after a mid-lethal dose of whole body irradiation.

Chaffee et al. (1966) and Phillips & Veath (1966) also reported increased kidney size in irradiated female mice but they did not attribute this finding to any histological changes.
Shah and Gadhia (1977) studied the effects of sub lethal (200r & 400r) total body gamma irradiation on kidney at different post-irradiation intervals like 1, 2, 4, 48 and 72 hour in pigeon and found that acid phosphatase activity increased in kidney tissue with both the doses of irradiation. Alkaline phosphatase showed decreased activity after 24 hour irradiation and succinate dehydrogenase showed a significant decrease in kidney tissue.

Peneyra & Jaenka (1985) studied the functional and morphological damage in the neonatally irradiated canine kidney. Whole body of Beagle dogs were exposed to 330 r to 60 Co gamma radiation at 4 days of age (IR4) to study the combined effects of direct radiation damage and nephron less, these findings indicate progressive glomerulosc-lerisis), PGS associated with perinatal renal irradiation result from direct radiation damage to deep cortical nephrons and compensatory functional changes occurring in response to loss of renal mass.

Jaenke and Angleton (1990) studied the perinatal radiation induced renal damage in the beagle dogs. The developing perinatal kidney is particularly sensitive to radiation.

The radiation induced changes by a single dose of 1000 r whole body exposure are reported to be intracellular edema, pycnosis and crenation, hyperaemia, cytoplasmic degranulation and vacuolization,
distortion of renal architecture and leucocytic infiltration (Purohit 1990).

Verma (1991) studied radiation induced changes after exposure to 10 Gy of gamma radiation in *Passer domesticus* (House sparrow) and the radiolesions observed in kidney were fatty degeneration, intracellular oedema, pycnotic nuclei, hyperaemia cytoplasmic degranulation and vacuolation, constriction of glomeruli, distortion of tubules, leucocytic infiltration etc.

Ding and Hu (1992) studied the effect of transfused marrow monocytes on masugi nephrities in rabbits with radiation injury. Sheep anti-rabbit glomerular basement membrane serum (NTS) was given to two groups of rabbits for establishing model of crescentic glomerulonephritis. Seven days after the inoculation, each rabbit received 800 rad whole body gamma-radiation in order to deplete the circulating leukocytes. Another 2 days later, all of the rabbits in group I received 10 cultivated rabbit marrow monocytes, while in group 2 nothing was given. 7 days after the administration of the cultivated cells, Bowman's capsules were broken with crescent formation. In rabbits of group 2, there were swelling of some epithelial and endothelial cells and crescents were scanty. The result proves that the macrophages are the main factors during the formation of crescent.

Yadav *et al.* (1994) studied the histological changes in the kidney of Swiss albino mice induced by fractionated doses of gamma rays. She
exposed the animals to three fractions of 1 or 2 Gy of gamma rays and effect was studied on day 1, 2, 3, 7, 15 and 30 after exposure. The kidney exhibited changes both in cortex and medulla regions. The various changes include waviness of the capsule, cytoplasmic degeneration, oedema and pycnotic nuclei. The radiolesions appeared on day-1 and progressed up to day-7 and signs of recovery were evident on day-15. The effects were found to be dose dependent.

The application of low dose radiation to an arterial ligation has the potential to subsequently reduce or eliminate restenosis caused by smooth muscle cell proliferation. Sufficient kidney irradiation causes a radiation nephropathy and often leads to renal failure. In order to evaluate the effect of low-dose irradiation on the kidney it was hypothesized that this particular therapy modifies renal injury in rats with renal ablation and subsequently slows the rate of the progression. For further clarification of the effect of irradiation at low doses, it was determined proliferating cell nuclear antigen (PCNA) and monocyte chemoattractant protein-1 (MCP-1) expression in remnant kidneys after low-dose radiation. Adult Wistar rats (n = 10) were studied during the two weeks after renal ablation. The left kidney was irradiated 24 hours after an operation in anaesthetised animals with 3 Grey in a single dose. Ablated rats without irradiation (n = 9) served as nephrectomized animals group. Rats without surgery and without radiation (n = 10) served as healthy controls. Renal damage was assessed using the following parameters: urine protein excretion rate (UprotV, mg/day), awake systolic blood pressure (SBP, mm Hg), serum creatinine (SCr,
micromol/l). The indirect immunofluorescence method was used for the detection of PCNA and MCP-1 expression. Glomerular and tubular immunostaining was scored semiquantitatively. Numerous PCNA positive cells and MCP-1 expression were present in the glomerulus and tubulointerstitium in nephrectomized rat kidneys. Low-dose radiation application was associated with a significant reduction in PCNA and low MCP-1 expression. It was observed that the application of low-dose irradiation has the potential to modify the progression of chronic renal failure in rats (Aunapuu et al., 2004).

**MERCURY**

The toxic effects of mercury are dependent on the compound to which exposure occurs and on the route of absorption. The kidney retains more mercury than any other organ in the body and the metal is in part excreted in the urine, principally by transfer through the tubular epithelium. Urinary excretion of mercury correlates with atmospheric concentration where group data are analyzed, but there can be considerable variation between individuals with similar exposure and in one individual from day to day. Estimation of urinary mercury concentration is of limited value in the diagnosis of mercurialism, as high excretion rates may be seen without clinical disorder, or mercurialism may be present when urinary excretion is low. Inorganic mercury compounds and a variety of organic mercurials have a pronounced diuretic effect in pharmacological dosage. Larger amounts of ingested inorganic mercury salts give rise to acute tubular necrosis.
which may result in oliguria or anuria. The prevalence of proteinuria is increased in workers exposed to mercury vapour, inorganic mercury and certain organic mercury compounds, an effect which appears to be related to glomerular damage. In some subjects the loss of protein through the glomerulus can be of such degree that the nephrotic syndrome may result. Indirect evidence suggests that an immunological mechanism may be involved. All workers exposed to mercury or its compounds should be screened regularly for proteinuria and removed from further exposure (Kazantzis, 1970).

Zalups (2000) studied that mercury is unique among the heavy metals in that it can exist in several physical and chemical forms, including elemental mercury, which is a liquid at room temperature. All forms of mercury have toxic effects in a number of organs, especially in the kidneys. Within the kidney, the pars recta of the proximal tubule is the most vulnerable segment of the nephron to the toxic effects of mercury. The biological and toxicological activity of mercurous and mercuric ions in the kidney can be defined largely by the molecular interactions that occur at critical nucleophilic sites in and around target cells. Because of the high bonding affinity between mercury and sulfur, there is particular interest in the interactions that occur between mercuric ions and the thiol group(s) of proteins, peptides and amino acids. Molecular interactions with sulfhydryl groups in molecules of albumin, metallothionein, glutathione, and cysteine have been implicated in mechanisms involved in the proximal tubular uptake, accumulation, transport, and toxicity of mercuric ions. In addition, the
susceptibility of target cells in the kidneys to the injurious effects of mercury is modified by a number of intracellular and extracellular factors relating to several thiol-containing molecules. These very factors are the theoretical basis for most of the currently employed therapeutic strategies. This review provides an update on the current body of knowledge regarding the molecular interactions that occur between mercury and various thiol-containing molecules with respect to the mechanisms involved in the renal cellular uptake, accumulation, elimination, and toxicity of mercury.

Ekawanti and Krisnayanti (2015) reported that mercury is a toxic metal with effects on human health ranging from acute to chronic in a very short time of exposure. Artisanal and small-scale gold mining (ASGM) is the main source of direct human exposure to mercury. Human exposure to mercury (Hg) can occur through both direct inhalation of mercury vapor and consumption of material taken from contaminated areas. To protect the health of ASGM workers and surrounding communities, a health assessment of mercury exposure and its effects is urgently needed. However, analysis of hair and urine samples as a proof test for mercury toxicity is very expensive. Therefore other tests must be considered to identify the first symptoms of mercury toxicity in miners and the surrounding community. It was aimed to determine the effects of mercury exposure on renal function along with the hematological parameters of gold miners and the community as a first indication of mercury exposure symptoms. The study was designed as a purposive field sampling study and was
conducted in three main villages in Sekotong District, West Lombok Regency, West Nusa Tenggara Province, Indonesia. The 100 subjects were miners that have been exposed to mercury for at least 5 years and their wives and children (non-miners) who lived around the gold processing area. Blood and urine samples were then obtained from the subjects. The miners and non-miners were questioned about their mercury exposure over the previous 5 years, duration of exposure, and how mercury was handled in their daily life. Blood and urine samples were collected at the time of the study; around 10 ml of urine and 0.1 ml of blood (2 drops) were collected per subject. In order to determine the parallel results between the blood-urine and hair results, hair from the miners was collected at a different time for analysis. The results showed that the subjects had low proteinuria, hemoglobin and hematocrit concentrations as a consequence of chronic mercury intoxication. This finding was parallel with results of high mercury concentrations in urine (>7 – 273.3 μg/l) and miners’ hair (>1 – 12.93 μg/g). Miners and non-miners in the exposure area were found to have proteinuria levels of more than 0.3 g/L. Proteinuria (≥0.3 g/L) was observed in 92.6% of miners and 72.4% of non-miners. It was concluded that urinalysis of proteinuria and hemoglobin values can be used as a screening test to detect renal impairment due to mercury intoxication.

Kidney injury molecule 1 (KIM-1, also known as TIM-1) is markedly upregulated in the proximal tubule after injury and is maladaptive when chronically expressed. It was determined that early
in the injury process, however, KIM-1 expression is antiinflammatory due to its mediation of phagocytic processes in tubule cells. Using various models of acute kidney injury (AKI) and mice expressing mutant forms of KIM-1, we demonstrated a mucin domain-dependent protective effect of epithelial KIM-1 expression that involves downregulation of innate immunity. Deletion of the mucin domain markedly impaired KIM-1-mediated phagocytic function, resulting in increased proinflammatory cytokine production, decreased antiinflammatory growth factor secretion by proximal epithelial cells, and a subsequent increase in tissue macrophages. Mice expressing KIM-1Δmucin had greater functional impairment, inflammatory responses, and mortality in response to ischemia- and cisplatin-induced AKI. Compared with primary renal proximal tubule cells isolated from KIM-1Δmucin mice, those from WT mice had reduced proinflammatory cytokine secretion and impaired macrophage activation. The antiinflammatory effect of KIM-1 expression was due to the interaction of KIM-1 with p85 and subsequent PI3K-dependent down modulation of NF-κB. Hence, KIM-1-mediated epithelial cell phagocytosis of apoptotic cells protects the kidney after acute injury by down regulating innate immunity and inflammation (Yang et al., 2015).

A study was conducted to summarize known findings related to exposure of aged and diseased kidneys to the environmentally relevant nephrotoxicant mercury by Bridges and Zalups (2017). Owing to advances in modern medicine, life expectancies are lengthening and leading to an increase in the population of older individuals. The aging
process leads to significant alterations in many organ systems, with the kidney being particularly susceptible to age-related changes. Within the kidney, aging leads to ultrastructural changes such as glomerular and tubular hypertrophy, glomerulosclerosis, and tubulointerstitial fibrosis, which may compromise renal plasma flow (RPF) and glomerular filtration rate (GFR). These alterations may reduce the functional reserve of the kidneys, making them more susceptible to pathological events when challenged or stressed, such as following exposure to nephrotoxicants. An important and prevalent environmental toxicant that induces nephrotoxic effects is mercury (Hg). Since exposure of normal kidneys to mercuric ions might induce glomerular and tubular injury, aged kidneys, which may not be functioning at full capacity, may be more sensitive to the effects of mercury than normal kidneys. Age-related renal changes and the effects of mercury in the kidney have been characterized separately. However, little is known regarding the influence of nephrotoxicants, such as mercury, on aged kidneys.

**COMBINED ACTION OF HEAVY METALS AND RADIATIONS:**

In today’s world where there are numerous physical, chemical and biological agents occurring in the environment, organisms will inevitably be exposed simultaneously to a combination of such agents. The interaction between radiation and other toxicants represents a field of immense potential importance as their total environmental burden may have greater effects than expected from the sum of their individual
impact. The total environmental burden is the result of a very complex network of interactions that may lead to greater effects than expected from the sum of the individual effects. Combined action of ionizing radiation and other agents are of potentially great importance as they may produce synergistic deleterious effect upon biological systems. According to UNSCEAR (1982) report, joint action of ionizing radiation and other agents are potentially great importance, because there are many occasions in our environment where interaction might occur.

A very little work has been carried out on the effects of metallic administration in combination with radiation. A supra-additive (synergistic) effect may be observed with a combination of radiation and metallic compounds.

Morphological development, cell proliferation and formation of micronuclei were used for assessment of risk after combined exposure to these metals and X-rays. They found that no conditions under which arsenic altered radiation risk; the effects were merely additive.

**CHEMOPROTECTION:**

The radiation response of any tissue depends upon the chemical nature of the target material and its environment. The radiation response can be reduced by modifying the environment of the target material chemically (chemical radioprotection).
Chemical radiation protection has a history of barely 50 years. The property of Sulphur compounds to protect against damage in complex chemical systems was recognized in the early 1940's. However, active research on the mammalian protection started about a decade later. The first demonstration that certain chemicals can reduce the X-ray induced damage was made by Dale (1942) in enzyme systems. But the first report on in vivo protection in mammals came in 1949, when Patt et al. published a study in which they observed that prior administration of cysteine, a naturally occurring amino acid, increased the survival of lethally irradiated rats and mice. This was followed by the discovery of more potent chemicals than cysteine in protecting against radiation induced mortality in laboratory mammals. Several compounds of different pharmacological activities were shown to have radioprotective property (Thomson, 1962), but none of them proved to be as good as the aminothiol chemicals, included under the cysteine-cysteamine group (Bacq and Alexander, 1961).

Thus, chemical protectors decrease the magnitude of such diverse responses like inactivation of enzymes, chromosomal aberration, gene mutation, acute radiation syndrome in mammals, erythema, epilation and delayed effects (Sterility and carcinogenesis). But every single protector is active in some organisms but not in others (Arena, 1971).

Early studies, were mostly concerned with understanding the structure and mechanism of action of the protectors. Later researches concentrated on the development of methods of reducing the toxicity of
the protector. These led to the screening of chemicals with less toxic but low protective activity as well as experimenting with various drug combinations.

Ghanekar and Koragaonkar (1972) reported the radioprotective effect of groundnut oil on skin of rabbit and Wistar rat after exposure to 70 KV and 220 KV of X-rays.

WR-2721 has been reported to selectively protect normal tissues and to concentrate in the liver. Utley et al. (1976a) observed an increased uptake of S\textsuperscript{35}, WR-2721 by liver and suggested the probability of its protection by this drug against radiation damage.

Khan (1980) studied the radioprotective effect of 2-MPG on liver of adult Swiss albino mice. He exposed mice to 2.5, 5.0 and 10.0 Gy of gamma radiation and found that the severity of damage, its tenure and the time of onset of recovery were dose dependent. He observed that the drug delays the appearance of pathological changes in the liver and an early recovery is observed in drug treated animals.

Purohit et al. (2001) observed modification of radiation induced biochemical changes in skin of Swiss albino mice by Liv.52. The animals were exposed to three fraction of 1 or 2 Gy of gamma rays at the intervals of 48 hrs. with and without Liv.52.

Purohit et al. (2002) also observed role of Liv.52 against radiation induced hepatic lesions in Swiss albino mice. They exposed the mice with three fractions of 1 or 2 Gy of gamma rays at the
intervals of 48 hrs. with (experimental) and without (control) Liv.52. The Liv.52 treated animals showed less severe radiolesions and fast recovery in comparison to control animals.

Dhir et al. (1990) evaluated extract of Phyllanthus *Emblica* fruit and ascorbic acid for protection against clastogenicity induced by lead (Pb) and Aluminum (Al) salts on mouse bone marrow chromosomes. Oral administration of Phyllanthus fruit extract (PFE) for 7 days before exposure to both metals by intraperitoneal injection increased the frequency of cell division and reduced the frequency of chromosome breaks significantly. Comparable doses of synthetic ascorbic acid (AA) were less effective and could protect against the effects of Al and only a low dose of Pb (10 mg/kg b.wt.). Ascorbic acid administered before treatment in mice given higher doses Pb (40 mg/kg b.wt.) enhanced the frequency of chromosome breaks, giving a synergistic effect. The higher protection afforded by PFE may be due to the combined action of ingredients, rather than to ascorbic acid alone.

Hari Kumar et. al. (2004) studied the radio protective effect of the fruit pulp of *E. officinalis* in Swiss albino mice. Mice were treated with 2.5 g/kg b. wt. of *Emblica* for 10 consecutive days before irradiation and exposed to a single dose of 700 rads (7Gy) of radiation after the last dose. One group was given *Emblica* continuously for another 15 days after irradiation. Changes in the total lucotocyte count, bone marrow viability haemoglobin were studied after whole body irradiation. Administration of *Emblica* significantly increased these
levels, which were lowered by irradiation. Animals were scarified at various time intervals after irradiation and the activities of the antioxidant enzyme catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPX) and glutathione-s-transferase (GST) and levels of glutathione were assayed in the blood. The damage to the cell membrane after whole body irradiation was studied by measuring the tissue lipid peroxide levels. Administration of *Emblica* significantly enhanced the activity of various antioxidant enzymes and GST as well as glutathione system in the blood. Treatment with *Emblica* also lowered the elevated levels of lipid peroxides in the serum.

Ganesh C. Jagetia (2006) studied the radioprotective potential of plants and herbs against the effects of ionizing radiation, the results obtained from *in vitro* and *in vivo* studies indicate that several botanicals such as *Gingko biloba, Cantella asiatica, Ocimum sanctum, Hippophae rhamnoides, Panax ginseng, Amaranthus paniculates, Mentha piperita, Zingiber officinale, Ageratum conzoides, Aphanamixis polystachya* protect against radiation induced lethality, lipid peroxidation and DNA damage.

A study was made on chemoprotective action of *Emblica officinalis* on skin carcinogenesis in mice by Sancheti *et al.* (2005). The inhibition of tumor incidences by fruit extract of this plant has been evaluated on two stage process of skin carcinogens in Swiss albino mice, induced by a single application of 7, 12-dimethyabenz (a) anthrecene and two weeks later, promoted by repeated application of
croton oil till the end of the experiment. The tumor yield, tumor incidence, tumor burden and cumulative number of pappillomas were found to be higher in the control as compared to experimental animals. The differences in the values of the results of experimental groups were statistically analysed and found to be significant in comparison to the control group (p<0.05).

Sharma and Purohit (2015) investigated the protective role of Liv.52 against radiation and cadmium induced haematological (RBC) changes in the Swiss albino mice. They exposed the animals with 3.0 and 6.0 Gy of gamma rays with or without cadmium chloride treatment. The more severe changes were observed after combined treatment which may be due to the synergistic effect. An early and fast recovery was also noticed in the drug treated group which showed the protection provided by the drug.

Purohit et al. (2014) studied therapeutic potential of Aloe vera against radiation and cadmium induced changes in the brain, kidney and liver of Swiss albino mice. Aloe barbadensis Mill, commonly known as Guar patha or Ghee patha, is perhaps the most important medicinal plant in the Indian traditional system of medicine, the Ayurveda. Several parts of the plant are used to treat a variety of diseases, but the most important is the leaf. It is rich in vitamins A, E and C (as antioxidant), minerals, enzymes, amino acids. Herbal plants are potential source of phytochemicals of pharmaceutical interest, act as antioxidants, anticarcinogenic and antimicrobial, preclinical studies.
Furthermore, experimental studies have reported that *Aloe* and some of its phytochemicals also exhibit radiomodulatory, chemoprotective, free radical scavenging and immunomodulatory activities. The exposure of living system to ionizing radiations causes a variety of damages to various systems due to generation of free radicals and reactive oxygen species. Cadmium is reported to be one of the most toxic elements in geological cycles. Various studies showed the prevention of radiation induced suppressions of immunity by *Aloe vera*. Having this unique properties *Aloe vera* could be used as protector against radiation and cadmium. Therefore, present study was planned to evaluate the protective efficacy of *Aloe vera* against radiation and cadmium induced changes in Swiss albino mice. For this purpose six to eight weeks old mice were selected and divided into seven groups on the basis of radiation, cadmium, combined treatment and drug treated. All biochemical parameters of the control groups were compared with the respective experimental groups. An increase in value of total proteins, glycogen, acid phosphatase, alkaline phosphatase and RNA was observed up to day 14 in non drug treated group and day 7 in the *Aloe vera* treated groups thereafter value declined up to day 28 without reaching the normal, whereas the value of cholesterol and DNA showed a decreasing trend. The biochemical finding indicated the drug treated section of living tissue (kidney, liver and brain) showed slightly / no degenerative changes. The drug treated groups demonstrating the ability of *Aloe vera* to inhibit oxidative stress thus preventing tissue injury.
Chakrawarti et al. (2015) observed histology and biomarkers: In mouse brain treated with radiation, cadmium and therapeutic agent *Aloe vera*. For this purpose, six to eight weeks old male Swiss albino mice (*Mus musculus*) were randomly divided into seven groups on the basis of radiation, cadmium, combined treatment and Aloe treated groups. The animals were sacrificed at each post-treatment intervals of 1, 2, 4, 7, 14 and 28 days. The brain were taken out and weighed to the analytical balance and fixed for 24 hours in alcoholic Bouin’s fixative. A pinch of lithium carbonate was added to remove excess picric acid in the fixative. Histological studies were carried out using the standard techniques of haematoxyline and eosin staining. After routine procedure of microtomy the sections were stained with Harris haematoxylene and eosin in alcohol, dehydrated in graded series of alcohol, cleared in xylene, mounted in DPX and examined microscopically. The histological changes observed were, pycnotic nuclei and crenated cells with condensation of nuclear material resulting into hyperchromatic cells. Hydrocephaly with enlarged lateral ventricles was also noted and corpus callosum was seen malformed. Thickened meninges and venous congestion were also noticed. In the irradiated brains cytoarchitectic layers were reduced in depth and showed some degree of intermixing of cells of various laminae. Hematoma was present between the cortex and medulla with numerous pycnotic and necrotic nuclei. Disarray of the cortical tissue with disorientation of cell processes was also evident. Damage in the cortex was noticed in the form of karyolysis, pycnosis and spongy
degeneration of the connective tissue with thickening of meninges. Dilation of blood vessel was also observed at certain places. After combined treatment of radiation and cadmium chloride synergistic changes were observed. These changes were less severe in the *Aloe vera* treated brain which may be due to the protection provided by drug.

*Hibiscus sabdariffa* L. has been used traditionally as herbal medicine and has been documented to have a broad range of therapeutic effects. The effects of chronic administration of aqueous extract of flowers of *Hibiscus sabdariffa* on the histology of the kidney and some biochemical indices of renal function in male Wistar rats have been investigated by Ukoha *et al.* (2015). Twenty Wistar rats were randomly divided into four groups of five rats each. The extract was administered orally in doses 200, 500, and 800 mg/kg body weight for 21 days. The kidney was harvested and processed histologically and blood samples were taken for biochemical assays. The histological results showed dose dependent pathological states and the biochemical analysis revealed a dose dependant variation in renal indices. These results suggest that chronic administration of aqueous extract of *Hibiscus sabdariffa* may be toxic to the kidney.

Ansar and Alghosoon (2016) evaluated the protective potential of diallylsulphide (DAS) against mercury-induced oxidative stress and antioxidant enzymatic alterations in spleen of rats. Rats were randomly divided into 4 groups of 6 rats each and exposed to mercuric chloride (*HgCl2*) (50 mg/kg, i.p.) and/or diallylsulphide (200 mg/kg/b.w) by gavage. Oxidative stress was evaluated in spleen by antioxidant
markers, viz, lipid per oxidation (LPO), superoxide dismutase (SOD), reduced glutathione (GSH), glutathione peroxidase (GPx) and catalase (CAT). Histomorphological changes in the spleen of rats were also compared between groups. Results: Oral administration of DAS at a concentration of 50 mg/kg daily showed a significant increase in GSH and GPx (p < 0.05), SOD and CAT (p < 0.05), as well as decreased LPO (p < 0.05) level in the spleen of rats as compared with HgCl₂ treated rats. Histopathology of spleen also showed that administration of DAS reduced the damage generated by HgCl₂ treatment. Conclusion: The results suggested that DAS may effectively normalize impaired antioxidant status in HgCl₂-induced oxidative stress. DAS has a protective effect against lipid peroxidation by scavenging free radicals and is thus capable of ameliorating mercury-induced changes in the spleen of adult Wistar rats.

Folate receptor (FR)-targeted radionuclide therapy using folate radioconjugates is of interest due to the expression of the FR in a variety of tumor types. The high renal accumulation of radiofolates presents, however, a risk of radionephropathy. A potential option to address this challenge would be to use radioprotectants, such as amifostine. Methods for early detection of kidney damage that—in this case—cannot be predicted based on dose estimations, would facilitate the development of novel therapies. The aim of this study was, therefore, to assess potentially changing levels of plasma and urine biomarkers and to determine DNA damage at an early stage after radiofolate application. The identification of an early indicator for renal
damage in mice would be useful since histological changes become apparent only several months after treatment. Mice were injected with different quantities of 177Lu-folate (10 MBq, 20 MBq and 30 MBq), resulting in mean absorbed kidney doses of ~23 Gy, ~46 Gy and ~69 Gy, respectively, followed by euthanasia two weeks (>85% of the mean renal radiation dose absorbed) or three months later. Whereas all investigated biomarkers remained unchanged, the number of γ-H2AX-positive nuclei in the renal cortex showed an evident dose-dependent increase as compared to control values two weeks after treatment. Comparison with the extent of kidney injury determined by histological changes five to eight months after administration of the same 177Lu-folate activities suggested that the quantitative assessment of double-strand breaks can be used as a biological indicator for long-term radiation effects in the kidneys. This method may, thus, enable faster assessment of radiopharmaceuticals and protective measures by preventing logistically challenging long-term investigations to detect kidney damage (Pellegrini et al., 2017).

*Moringa oleifera* (Shaijan)

*Moringa oleifera* is a fast-growing tree native to South Asia and now found throughout the tropics. Its leaves have been used as part of traditional medicine for centuries, and the Ayurvedic system of medicine associates it with the cure or prevention of about 300 diseases. Moringa, sometimes described as the “miracle tree,” “drumstick tree,” or “horseradish tree,” has small, rounded leaves that
are packed with an incredible amount of nutrition: protein, calcium, beta carotene, vitamin C, potassium… you name it, moringa’s got it. No wonder it’s been used medicinally (and as a food source) for at least 4,000 years.

The fact that moringa grows rapidly and easily makes it especially appealing for impoverished areas, and it’s been used successfully for boosting nutritional intake in Malawi, Senegal, and India. In these areas, moringa may be the most nutritious food locally available, and it can be harvested year-round.

*Moringa oleifera* is a tree that is sometimes called the Tree of Life or a Miracle Tree, but rather than this being in reference to its potential medicinal usage this is actually referring to how it is a very valuable food crop (it is drought resistant, grows very fast, and is highly nutritive) and even beyond food it serves many benefits in third world countries such as having an ability to be used for some crafts (due to being a tree) and cleaning water.

For usage as a supplement, *Moringa oleifera* is recommended mostly as being a highly nutritious antioxidant. While it is indeed nutritious, supplemental dosages are too low to acquire adequate nutrition from and this claim is not relevant; it is a relatively potent antioxidant, and while it seems to be less potent than other herbs when tested outside of a living system it does appear to be quite potent when tested in living models. The reason for the increased potency in living models is not known (although it is possible that it can induce genetic
transcription similar to Sulforaphane since the bioactives are similar in structure), but the antioxidant properties seem to underlie the vast majority of benefits associated with this supplement.

There are also anti-inflammatory effects that, while less studies, seem to be quite effective; one of the bioactives, RBITC, is effective in suppressing macrophage activation in the nanomolar range which is worth some future research into. Beyond that, there does appear to be a nice anti-diabetic effect that has gone some very preliminary human testing which suggests that this plant may benefit pancreatic function and reduce blood glucose secondary to that.

While both the antioxidant and anti-inflammatory properties are somewhat interesting, until the exact mechanisms and relative potency to some other antioxidants or anti-inflammatories are tested it is hard to recommend this supplement over other options.

Now, despite the plant being referred to as 'nontoxic' this does not appear to be the case overall. While supplemental dosages listed below appear to be safe from all tested toxicity a relatively small increase (3-4 times the recommended does) is known to cause genotoxic damage and may promote cancer formation whereas doses higher than that cause overt organ damage (mostly liver and kidneys); this effect is seen with the seeds while toxicity of the leaves seems to be a lesser concern. Beyond that, very reasonable supplemental dosages
appear to be able to induce abortions in pregnant rats and thus supplementation is contraindicated (not advised) in pregnant women.

The safety of an aqueous leaf extract given orally to rats at doses of 400, 800, 1600, and 2000 mg/kg body weight was examined (Adedapo et al., 2009). The treatment was either an acute single dose or given daily for 21 days except the highest dose. Various parameters were assessed including blood cell counts and serum enzyme levels. The authors concluded that consumption of *M. oleifera* leaves at doses of up to 2000 mg/kg were safe. A dose-dependent decrease in body weights of the rats occurred over the 21 days of the study.

Ambi et al. (2011) divided 24 rats into four groups and fed varying amounts of *M. oleifera* powdered leaves mixed with standard livestock feed (25%, 50%, 75%, and control) for 93 days. Total amount of *M. oleifera* leaves consumed was not quantified. Following the experimental period, some organs of the treated animals had observable microscopic lesions with the 75% group developed necrosis of hepatic cells, splenic blood vessels, and neuronal glial cells. The control animals had no observable microscopic lesions in all organs examined. No photomicrographs of any tissues were provided. The amounts of leaves consumed, although not quantified by the authors, greatly exceeded doses that would be typically used in either rats or humans. For example, if the rats consumed an average of 15–20 g of chow per day, even at the low dose of 25% of the chow, the daily dose would be
approximately 15–20g of leaves per kilogram for an adult rat, which would equate to 195–260g for an 80-kg human.

The genotoxicity of an aqueous *M. oleifera* seed extract was assessed using three separate assay systems including the Ames assay (Rolim *et al*., 2011). The seed extract was not genotoxic without metabolic activation, and did not pose a risk to human health. The effect of a hexane extract of *M. oleifera* leaves on reproductive organs of male rats was examined (Cajuday and Pocsidio, 2010). The extract was given orally at doses of 17, 170, and 1700mg/kg body weight for 21 days. A dose-dependent increase in testis and epididymis weights, in seminiferous tubule diameter, and epididymal epithelium thickness without change in plasma gonadotropin levels was observed. The authors concluded that the changes were associated with an increase in spermatogenesis.

Asare *et al*. (2012) examined the potential toxicity of an aqueous leaf extract of *M. oleifera* in several different experimental systems. In one set of experiments, human peripheral blood mononuclear cells were exposed *in vitro* to graded doses of the extract and cytotoxicity was assessed. Cytotoxicity occurred at 20 mg/kg, a concentration not achievable by oral ingestion. In another set of experiments, rats were given 1000 and 3000 mg/kg of the extract, and the animals were assessed for up to 14 days. The *M. oleifera* leaf extract was shown to be genotoxic based on blood cell analysis at the 3000 mg/kg dose, a dose that greatly exceeds commonly used doses. A dose of 1000 mg/kg
was deemed safe and did not produce genotoxicity when given to rats, a dose still in excess of commonly used doses.

The toxicity of an aqueous extract of *M. oleifera* leaves has also been evaluated in mice (Awodele *et al.*, 2012). In an acute study, mice were administered the extract at up to 6400 mg/kg orally and 1500 mg/kg intraperitoneally. In a subchronic study, mice received 250, 500, and 1500 mg/kg orally for 60 days. The lethal dose of 50% LD50 was estimated to be 1585 mg/kg. No significant effects were observed with respect to hematological or biochemical parameters or sperm quality. A high degree of safety was observed on oral administration.

*Moringa oleifera* Lam. is a tree that grows widely in many tropical and subtropical countries. It is grown commercially in India, Africa, South and Central America, Mexico, Hawaii, and throughout Asia and Southeast Asia. It is known as the drumstick tree based on the appearance of its immature seed pods, the horseradish tree based on the taste of ground root preparations, and the ben oil tree from seed-derived oils. In some areas, immature seed pods are eaten, while the leaves are widely used as a basic food because of their high nutrition content (Thurber and Fahey, 2009; Mbikay, 2012; Razis *et al.*, 2014). No human clinical trials have been conducted looking at the efficacy of *M. oleifera* for treating under nutrition.

Seeds, leaves, oil, sap, bark, roots, and flowers are widely used in traditional medicine. *Moringa* leaves have been characterized to
contain a desirable nutritional balance, containing vitamins, minerals, amino acids, and fatty acids (Moyo et al., 2011; Teixeira et al., 2014; Razis et al., 2014). Additionally, the leaves are reported to contain various types of antioxidant compounds such as ascorbic acid, flavonoids, phenolics, and carotenoids (Alhakmani et al., 2013; Vongsak et al., 2014). According to several commentaries (Anwar et al., 2007; Mbikay, 2012; Razis et al., 2014), various preparations of *M. oleifera* are used for their antiinflammatory, antihypertensive, diuretic, antimicrobial, antioxidant, antidiabetic, antihyperlipidemic, antineoplastic, antipyretic, antiulcer, cardioprotectant, and hepatoprotectant activities. The therapeutic potential of *M. oleifera* leaves in treating hyperglycemia and dyslipidemia was reviewed by Mbikay (2012). Razis et al. (2014) summarized potential health benefits of *M. oleifera*, focusing on their nutritional content as well as antioxidant and antimicrobial characteristics.

Paul and Didia (2012) investigated the effect(s) of methanol extract of *M. oleifera* root on the histo-architecture of the liver and kidney of 24 guinea-pigs. Experimental conditions included daily intraperitoneal injections of the root extract at doses of 3.6, 4.6, and 7.0 mg/kg, and control for 3 weeks. Histological sections of all treated groups had ballooning degeneration of the liver, suggesting time-dependent hepatotoxicity rather than a dose-dependent response. Examination of the kidneys, demonstrated mild tubular damage and interstitial inflammation in the 4.6mg/kg group, while the 7.0 mg/kg
group had infiltration of the interstitium by inflammatory cells and amorphous eosinophilic materials. No information was provided regarding extract composition or degree of concentration. The results of this study cannot be compared or equated with studies involving aqueous extracts of leaves. This study involved a methanol extract of roots, which was given intraperitoneally and not orally.

Sharma et al. (2012) studied the renoprotective effect of *Moringa oleifera* pods in 7, 12 dimethylbenz[a] antracene- exposed mice. They investigated the potential of hydroethanolic extract of *Moringa oleifera* (MOHE) against 7, 12-dimethylbenz[a]anthracene (DMBA)-induced toxicity in male Swiss albino mice. Experimental mice were respectively pre-treated with 200 and 400 mg/kg of MOHE, and 0.5% and 1% of butylated hydroxyanisole (BHA) for two weeks prior to the administration of 15 mg/kg of DMBA, respectively. Levels of xenobiotic metabolizing enzymes such as cytochrome (Cyt) P450 and Cyt b5, activities of reduced glutathione (GSH) and glutathione-S-transferase (GST) and renal aspartate amino transaminase (AST), alanine amino transaminase (ALT) and alkaline phosphatase (ALP), and content of protein and total cholesterol were measured to determine the nephrotoxicity caused by DMBA and to elucidate the ameliorating role of *M. oleifera*. Single oral administration of 15 mg/kg of DMBA resulted in significant increases in Cyt P450 and Cyt b5 (P<0.01). The toxic effect of DMBA was justified by the significant decreases in the activities of GSH and GST in renal tissues (P<0.05). The levels of renal
AST, ALT and ALP and protein content which are indicative of renocellular damage were also found decreased along with significant increase in total cholesterol content in DMBA-treated mice (P<0.01). The DMBA-induced alterations in the tissues were significantly reversed after pre-treatment with 200 and 400 mg/kg of MOHE orally for 14 d (P<0.01). It was concluded that the the effects of MOHE in enhancing the levels of antioxidants and enhancing the levels of biochemical assays in DMBA-induced carcinogenesis are by reducing the formation of free radicals. The study rationalized the ethnomedicinal use of *M. oleifera* for the protection against nephrotoxicity induced by chemical carcinogens.

The toxicological effects associated with consumption of 50, 100, 200, or 400 mg/kg of methanol extract of *M. oleifera* for 8 weeks was performed in 30 rats (Oyagbemi *et al*., 2013). The extract was a 30:1 concentration. All experimental animals that received *M. oleifera* had a significant increase in body weight in a dose-dependent manner, contrary to what is observed with an aqueous extract (Adedapo *et al*., 2009). Rats that received *M. oleifera* at 200 and 400 mg/kg showed a significant increase in serum alanine aminotransferase, aspartate aminotransferase, blood urea nitrogen, and creatinine. It should be noted that the extract was prepared with methanol and not water. The 30:1 concentration of the methanol extract at a dose of 400 mg/kg would be equivalent to 12 g of leaves per kilogram, a very unrealistic dose. The composition of the extract was not reported, and it is not
clear how the composition of the methanol extract relates to the composition of aqueous extracts, which are commonly used.

Bakre et al. (2013) determined that the lethal dose of 50% of an orally administered ethanol extract of *M. oleifera* leaves in mice was greater than 6.4g/kg. The dietary effects of *M. oleifera* leaves as a dietary supplement for liver function were performed by Zvinorova et al. (2014). Thirty-two weanling rats were randomly assigned to diets of normal rat feed fed at 20% and 14% of body mass, or *Moringa*-supplemented feeds fed at 20% and 14% of body mass for 5 weeks. *Moringa* supplementation did not affect blood metabolite concentrations, liver glycogen, or lipid storage.

The potential toxicological effects of a single oral dose of 5000 mg/kg of an aqueous *M. oleifera* extract as well as oral doses of up to 1000 mg/kg of the same extract for 14 days on rats were examined (Asiedu-Gyekye et al., 2014). The authors noted that no overt adverse reactions were observed at these doses, and no histopathological findings were found. Small but statistically significant dose-dependent increases in several liver enzymes were observed. A dose of 1000mg/kg in a rat is equivalent to over 30 times a typical 400 mg dose of an aqueous extract in an 80-kg human.

*Moringa oleifera* (Sajna in India) is considered to be a very popular vegetable among the Indian and African continent. Among its different parts, the leaf holds the best nutritional and medicinal
properties. The present study was aimed to evaluate the protective action of *Moringa oleifera* leaf extract (*MoLE*) against oxidative stress induced DNA damage. To that end, the hydroxyl radical mediated DNA damage by Fenton reaction and hydrogen peroxide (H\textsubscript{2}O\textsubscript{2}) mediated DNA damage by comet assay were employed. Further, lipid per oxidation (LPO) was assayed to find whether *MoLE* can prevent the subsequent membrane damage. The extract prevented hydroxyl radical induced DNA damage as well as H\textsubscript{2}O\textsubscript{2} mediated comet formation as determined by fluorescence microscopy. In addition, *MoLE* inhibits the LPO level by about 30% at 100 μg/ml concentrations. *MoLE* also inhibits the Topoisomerase I activity which is one of enzymes responsible for DNA metabolism. *MoLE* shows high polyphenol content (50 mg polyphenols/g dry leaf), strong reducing power, high metal chelating capacity and DPPH (2, 2-diphenyl-2-picryl hydrazyl) radical scavenging activity. All these parameters can immediately be correlated to its DNA protection efficacy. The present study, first time to our knowledge indicates that *MoLE* possesses significant DNA protective activity and hence suggested for the potential human consumption against oxidative DNA damage and cell damage (Sikder *et al.*, 2013).

For the sake of completeness, several studies involving *M. oleifera* seeds and roots will be described, although the results cannot be directly compared or equated with studies involving leaves. Cytotoxicity of an aqueous extract of *M. oleifera* seeds was evaluated
by Araújo et al. (2013). Following 14 days of the extract administration (500 and 2000 mg/kg) in mice, no signs of systemic toxicity were observed, and all the animals survived. There were no changes in organ indices between treatment and control groups. Small but insignificant changes were observed in erythrocytes, platelets, hemoglobin, and hematocrit. All values remained within the normal range.

A methanol extract of seeds of *M. oleifera* were screened phytochemically for chemical components and used for acute and subacute toxicity studies in rats (Ajibade et al., 2013). The phytochemical screening revealed the presence of saponins, tannins, terpenes, alkaloids, flavonoids, carbohydrates, and cardiac glycosides but the absence of anthraquinones. Although signs of acute toxicity were observed at an extract dose of 4000 mg/kg, mortality was recorded at 5000 mg/kg. No adverse effects were observed at concentrations lower than 3000 mg/kg. It was concluded that methanol extracts of seeds of *M. oleifera* are safe for nutritional use.

Eshak and Osman (2013) studied the role of *Moringa oleifera* as a protector in male rats against oxidative stress induced by gamma irradiation. Fifty male albino rats were randomly divided into five groups. Group I; animals without any treatment, as control. Group II; animals exposed to 4Gy, low dose of gamma irradiation. Group III; animals exposed to 6Gy, high dose of gamma irradiation. Group IV; animals exposed to 4Gy gamma irradiation then gavage with 50 mg/kg body weight daily of *Moringa oleifera* water extract for one month.
Group VI; animals exposed to 6Gy gamma irradiation then gavaged with 50 mg/kg body weight daily of *Moringa oleifera* water extract for one month. The evaluated haematological parameters were, iron, vitamin B12 and folic acid. AST, ALT, ALK.Ph. and malondialdehyde (MDA) activity and level were analyzed respectively. The genetic parameters were evaluated by means of DNA fragmentation, micronucleus test and comet assay. The results revealed a significant decrease in RBCs, Hb, Ht% and platelets, however, MCV and MCH increased significantly after exposure to low and high doses of irradiation. *Moringa oleifera* treatment ameliorated these effects, especially in the low gamma irradiation dose and improved the anemia. Gamma irradiation also decreased significantly: WBCs, lymphocytes, monocytes, neutrophils, esinophiles and basophiles. The decrease was more pronounced in the 6Gy high dose than 4Gy low dose. After treatment with *Moringa oleifera* at the low or high doses, lymphocytes, monocytes and basophiles counts increased more with the low dose than with the high dose. Iron, vitamin B12 and folic acid decreased significantly after exposure to gamma irradiation with 4Gy and 6Gy doses. After *Moringa oleifera* treatment, the low dose group restored their levels close to the control values. The AST, ALT and ALK. Ph. activities increased proportionally with the gamma irradiation dose. *Moringa oleifera* treatment decreased these levels below the normal control levels in the 4Gy dose. However, at the 6 Gy dose, ALK.Ph. activity still slightly increased, while AST and ALT levels approached the control levels. *Moringa oleifera* recovered the hepatic damage.
Malondialdehyde (MDA) increased with low and high doses of irradiation and *Moringa oleifera* decreased these effects by lowering oxidative damage. The genotoxicity study revealed that exposure of male rats to gamma irradiation increased the DNA fragmentation, frequency of the micronucleated polychromatic erythrocytes (MnPCEs) formation and the DNA damage. These effects were more pronounced with the high dose of irradiation than with the low dose. However, the DNA fragmentation, frequency of the MnPCEs formation and the DNA damage decreased after the treatment of irradiated male rats with *Moringa oleifera* leaf extract.

Oyagbemi *et al.* (2013) investigated toxicological effects associated with prolonged consumption of *Moringa oleifera* leaves as a beverage. Thirty rats were used in this study. They were grouped into five groups of six rats. Rats in group I received 2 mL/kg body weight (b.w.) of corn oil (vehicle). Animals in groups II, III, IV and V received 50, 100, 200 and 400 mg/kg b.w. of methanolic extract of *M. oleifera* for eight weeks. Serum collected was analyzed for alanine aminotransferase (ALT), aspartate aminotransferase (AST), total protein, albumin, globulin, blood urea nitrogen (BUN) and creatinine. There was a significant (p<0.05) increase in serum total protein and globulin in a dose-dependent manner. Rats that received *Moringa* at 200 and 400 mg/kg b.w. showed a significant (p<0.05) increase in serum ALT, AST, BUN and creatinine which pointed to hepatic and kidney damage. All experimental animals that received *Moringa* had a significant (p<0.05) increase in body weight in a dose-dependent
manner. This study therefore confirms for the first time that chronic administration of *M. oleifera* leaves might predispose to hepatic and kidney damage.

In summary, based on human, animal, and *in vitro* studies, and the extrapolation of results from animal studies to humans, various preparations of *M. oleifera* leaves including aqueous extracts appear to be exceedingly safe at the doses and in the amounts commonly utilized.

Seriki *et al.* (2015) examined the blood cell (White and Red cell) counts of Wistar rats fed with ethanolic extract of *Moringa oleifera* leaves. For the study, Wistar rats (18) were divided into three groups of 6 each: Group 1 served as control (not given extract), Group 2 was fed with 200mg per kg (of body weight) of the leaf extract for 15 days, and Group 3 was fed with 300mg per kg (of body weight) of the leaf extract for the same duration. **The results** showed a significant increase (P<0.05) in both red and white blood cell counts of rats in groups 2 and 3 as compared to the ones in the control group (group 1) in both low and high doses. They concluded that 200 mg per kg and 300mg per kg of *Moringa oleifera* leave extract contain active ingredients required for the formation and maturation of blood cells (red and white blood cells), hence the increase in the blood cells counts.

Protective effect of *Moringa oleifera* leaf extract against radiation induced lipid peroxidation has been investigated by Sinha *et al.* (2012). Swiss albino mice, selected from an inbred colony, were administered
with *Moringa* extract (300 mg/kg body wt.) for 15 days before exposing to a single dose of 5.0 Gy of gamma radiation. After treatments, animals were necropsied at different post-irradiation intervals (days 1, 7 and 15) and hepatic lipid peroxidation and reduced glutathione (GSH) contents were estimated to observe the relative changed due to irradiation and its possible amelioration by *Moringa* extract. It was observed that, *Moringa* treatment restored GSH in liver and prevented radiation induced augmentation in hepatic lipid peroxidation. Phytochemical analysis showed that Moringa possess various phytochemicals such as ascorbic acid, phenolics (catechin, epicatechin, ferulic acid, ellagic acid, myricetin) etc., which may play the key role in prevention of hepatic lipid peroxidation by scavenging radiation induced free radicals.

The leaf of *Moringa oleifera* has a wide range of beneficial effects which was predicted in Indian system of medicine (Ayurveda and Unani). *Moringa oleifera* leaves have been reported to possess immunomodulatory (Gupta *et al.*, 2010) and wound healing (Rathi *et al.*, 2006) properties. It was also reported to prevent gastric ulceration (Debnath and Guha, 2007). There are reports that the leaf extracts prepared using either by methanol or by ethanol inhibit microsomal lipid peroxidation (Sidduraju and Becker, 2003). *Moringa* also showed radioprotective potential as the leaf extract protects bone marrow chromosomes against radiation induced damage (Rao *et al.*, 2001). Therefore, it has been hypothesized that leaf extract of *Moringa oleifera* may inhibit the radiation induced lipid peroxidation.
Liver is the most metabolically active organ and it reflects any systemic derangement upon ionizing radiation. After radiation exposure liver is affected significantly (Kumar et al., 2005 and Bhatia and Jain., 2004). It has been reported that extract from *Moringa oleifera* is able to protect the liver against acetaminophen (Fakurazi et al., 2008), carbon tetrachloride induced damage (Selvakumar and Natarajan, 2008). However, the effect of *Moringa* leaf against radiation induced alteration in liver has not been reported. Hence, in the present investigation an attempt has been made to study the protective effect of *Moringa oleifera* leaf extract in radiation induced oxidative insult. The protective effect was evaluated in terms of lipid peroxidation using liver as an experimental model. The study also aimed at identifying phytochemicals present in *Moringa oleifera* leaf extract which may be responsible for the prevention of *in-vivo* lipid peroxidation.

*Moringa* flowers contain a well recognized flavonoid (Quercetin), which may be responsible for its potent hepatoprotective activity (Ruckmani et al., 1998; Selvakumar and Natarajan, 2008). In a recent study evaluating the effect of *Moringa oleifera* seed extract on liver fibrosis, it was found that *Moringa* seed extract has the ability to subside liver fibrosis. This study involved CCl₄ induced liver fibrosis and concurrent administration of *Moringa oleifera* seed extract. *Moringa oleifera* seed extract control the elevation of serum aminotransferase activities and globulin level induced by CCl₄. Moreover, immunohistochemical studies also showed that *Moringa oleifera* reduces liver fibrosis (Hamza, 2010).
The extract of *Moringa* leaves has been shown to have potent antioxidant action in vivo (Sreelatha and Padma, 2011). Because this part of plant when compared to other parts, it was found that it is high significant source of protein, β-carotene, vitamins A,B,C and E, riboflavin, nicotinic acid, folic acid, pyridoxine, amino acids, minerals and various phenolic compounds (Khalafalla *et al*., 2010). Also, *Moringa oleifera* leaves were used as nutritional supplement and growth promoters (Sanchez *et al*., 2006). The extract of this part of the plant has been investigated to be a protective or therapeutic agent against various abnormal conditions. Ethanolic extract of leaves has shown antimicrobial activity (Nepolean *et al*., 2009).

Radio protective effect of leaves has also been established where in radiation-induced chromosomal aberrations and micronuclei were suppressed by pre-treatment with methanolic extract. Moreover, *M. oleifera* leaves aquous extract was observed to have a therapeutic action against radiation hazards through enhancing of liver enzyme activities (AST, ALT and ALK), decreasing the malondialdehyde (MDA), and reduction of genetic alterations (micronuclei and DNA damage) in irradiated rats by gamma irradiation (Eshak and Osman, 2013). Furthermore, ethanolic extract of *M. oleifera* leaves possessed antigenotoxic phyto constituents in mice, the high percentages of micronuclei and DNA damage induced by cyclophosphamamide were minimized in animals pre-dosed with the extract (Sathya *et al*., 2010).

Meena (2013) investigated radioprotective effect of *Moringa oleifera* (Lam.) seed and *Acorus calamus* (Linn.) rhizome extract on
mice. The animals were exposed with 8.0 Gy of gamma radiations with or without *Moringa* administration. The study showed that DNA and acid phosphatase content was higher at the subsequent interval but did not attain the normal value, while RNA and alkaline phosphatase reached the maximum value on 4\(^{th}\) day and then declined towards normal. The alterations in the value were less severe in the drug treated groups showing protection provided by the drug.

Ionizing radiation generates reactive oxygen species that induce oxidative stress, which generates free radicals, which associated with many degenerative diseases. The present study has been designed to evaluate the ameliorative role of aqueous *Moringa oleifera* leaf extract (MO) against \(\gamma\)-radiation (IRR)-induced oxidative stress in hepatic and renal tissues in rats. Twenty four male albino rats were divided into four groups, (1) control group injected with the vehicle, (2) MO treated group, (3) IRR group, (4)MO/ IRR treated group. Biochemical and ultra structural examinations were utilized for evaluation of the oxidative stress, hepatotoxicity and nephrotoxicity. IRR (6Gy) caused a significant increase in liver and kidney malondialdehyde (MDA) and total nitrate/nitrite (NO (x)) levels and significant decrease in superoxide dismutase (SOD), catalase (CAT) activities and glutathione (GSH) content. The activities of alanine aminotransferase (ALT) and aspartate aminotransferase (AST), and the levels of urea nitrogen and creatinine in serum were increased. Administration of MO (300 mg/kg, i.p.) for 15 days prior to IRR ameliorated the hepatotoxicity and nephrotoxicity induced by IRR. Ultra structural examination of liver
and kidney tissues confirmed the biochemical data. The present results revealed that MO has a protective effect against IRR-induced hepatotoxicity and nephrotoxicity through its free radical scavenging activity and enhancement of the antioxidant defense mechanisms (Heba et al., 2014).

An attempt has been made to investigate the effects of ethanolic *Moringa oleifera* leaf extract on the histology of vital body tissues. The rationale is that histological observations would provide a more reliable and consistent picture of the effects produced by the interactions of the phytochemicals with the body cells and tissue. It may be helpful in observing the possible toxicological effects on body tissues or on the other hand, the positive effects on the body tissues. A total of twelve Wistar rats (n=12) were used for the investigation; divided in two groups of Control (A) and Treated (B). A daily dosage of 200mg/kg body weight of ethanolic *moringa* leaf extract was administered orally to the treated Group B for 28 days. Analysis of each tissue’s histomorphology, general histo-architecture and cytological structures was critically done. The basis of analyses and inferences was clearly defined: whether *Moringa oleifera* leaf extract produced any observable deleterious effects on the toxicological evaluation of tissue; or whether its effects would improve the tissue’s histological architecture especially in manners that can produce improvement in physiological conditions of the individual tissue or general body health. Extract produced positive effects in the liver (Owolabi and Ogunnaike, 2014).
The protective and therapeutic effect of *Moringa oleifera* leaf extract (MOLE) against carbon tetrachloride (CCL₄)-induced genotoxicity, hemotoxicity and hepatotoxicity in rats has been evaluated by Eshak et al., 2015. Male albino rats of eleven groups (eight animals each) were used in the study. The animal groups included negative control; control of olive oil; positive control (received CCL₄ in olive oil for 12 weeks); Groups 4-7 received CCL₄ in olive oil plus MOLE at doses of 1.3, 2.0, 2.6 and 4.0 g/kg (used as a protective agent) for 12 weeks; Groups 8-11 received MOLE alone (as a therapeutic agent) at the same doses for three weeks after cessation (12 weeks) of CCL₄ treatment. Molecular genetics, hematological, histopathological and histochemical studies were conducted. Genetic results showed that the administration of CCL₄ caused a high significant increase of DNA damage in lymphocyte cells and significant elevation of the expression of CYP1A2 and CYP2B1 genes in liver tissue as compared to control. Also hematological findings revealed that CCL₄ treatment significantly reduced Hb level and RBCs count, whereas it significantly increased the WBCs count in respect to normal control. Histopathological examination documented that CCL₄ produced massive damage to liver tissue in the form of excessive fibrosis, cellular infiltration and vacuolar degeneration of hepatocytes. MOLE treatment (as a protective or therapeutic agent) was able to significantly reduce the DNA damage and significantly inhibit the up-regulation of CYP1A2 and CYP2B1 genes expression induced by CCL₄. It also significantly improved the hematological parameters,
where the abnormal changes in Hb level, RBCs count and WBCs count induced by CCL₄ had been minimized. Moreover, the histopathological results revealed that the damaging effect of CCL₄ on hepatic tissue was clearly reduced by using MOLE treatment. Histochemical findings confirmed the histopathological results, where the DNA study indicated that MOLE treatment ameliorated the DNA content in examined cells and gave DNA values better than those of animals group treated with CCL₄ alone. While the CCL₄ group showed decrease of DNA values (hypoploidy). All the results were dose dependent. But better results were obtained by using MOLE as a therapeutic agent, especially the treatment with the highest dose 4.0 g/kg, in which the rate of DNA damage, the over expression especially of CYP1A2 gene, hematological changes and the massive damage in liver tissue as well as the abnormal histochemical parameters reverted nearly to the normal values. It was concluded that MOLE is able to significantly alleviate the oxidative stress induced by CCL₄ in rats., These results revealed that *Moringa oleifera* has therapeutic effect in curing some health problems associated with toxification status (as a result of CCL₄ treatment) and this was established by its positive effect on some of molecular genetics, hematological, histopathological and histochemical parameters of the experimental animals.

*Moringa oleifera* belongs to family of moringaceae and is considered as one of the world’s most useful trees, as almost every part of the plant can be used for either as food, or therapeutic purposes. Fatty diet is a significant factor in the pathogenesis of non-alcoholic
fatty Liver disease (NAFLD). The study was designed to examine histological effects of aqueous extract of *Moringa oleifera* on the liver tissues of wistar rat fed with high fat diet. Twenty five adult rats were divided into five groups of five animals each. While group A received distilled water daily only, groups Band C received aqueous extract of *Moringa oleifera* at doses of 200 mg/kg body weight and fat high diet (30% w/w of the total mash feed) respectively for a duration of seventy days. Others (groups D and E) received aqueous extract of *Moringa oleifera* at doses of 200 mg/kg body weight and fat high diet for a duration of seventy days. Histology of the liver of the rats fed with high fat diet exhibited significant changes in the architecture of liver tissue. The changes include micro and macro vascular steatosis, increased fatty infiltration, inflammation, sinusoidal dilation, degeneration of veins and vacuolization as compared to normal liver histology. Treatment with 200 mg/kg extract of *Moringa oleifera* significantly attenuated these effects imposed by high fat diet as compared to the control group. Therefore demonstrated that daily administration of *Moringa oleifera* leaves extract to rats for a period of 70 days may reverse the formation of hepatic steatosis in nonalcoholic fatty liver disease. Keywords; Vascular steatosis, non-alcoholic fatty liver disease (Obayuwana et al., 2016).

Abarikwu et al. (2017) studied protective effect of *Moringa oleifera* oil against mercuric chloride induced hepato- and nephrotoxicity in rats. They reported that various parts of the *Moringa oleifera* (*M. oleifera*) tree are widely accepted to have ameliorative effects
against metal toxicity. For the study, *M. oleifera* oil (MO) was tested against mercuric chloride induced tissue pathologies and oxidative stress. Male Wistar rats were administered MO (1.798 mg/kg p.o.) or HgCl$_2$ (5 mg/kg body wt) alone or in combination (5 mg/kg HgCl$_2$+1.798 mg/kg MO p.o.) three times per week for 21 days. After exposure and treatment periods, rats were sacrificed; blood collected and the oxidative status of the liver and kidney homogenates were evaluated. In the liver, malondialdehyde (MDA) level, glutathione (GSH), and superoxide dismutase (SOD) activities were higher whereas catalase (CAT) activity was lower in the HgCl$_2$ group than in the control group. In the kidney, MDA level, SOD, and CAT activities were higher whereas GSH activity was unchanged in the HgCl$_2$ group compared to the control group. In the liver, MDA level, SOD, and CAT activities were lower in the HgCl$_2$+MO group than in the HgCl$_2$ group. In the kidney, MDA level, SOD and CAT activities were lower in the HgCl$_2$+MO than in the HgCl$_2$ group. Furthermore, Hg-induced increases in creatinine and bilirubin levels as well as the increase in γ-glutamyl transferase, lactate dehydrogenase, and alkaline phosphatase activities were attenuated in the combine exposure group and the animals showed improvement in the histology of the liver and kidney. It was included that MO decreased the negative effects of Hg-induced oxidative stress in rats.

Purohit *et al.* (2017) studied protective influence of *Moringa oleifera* against radiation and mercury induced hepatotoxicity in mice. Radiation induced damage and lethality to the normal tissues can be
partially reduced by the use of radio-protectors that lower down the damaging effects of radiation. In recent years, extensive research work has been carried out on chemical protection against radiation and heavy metals induced toxicity. Several synthetic chemicals have been tested for their radio-protective action in mammals but their practical applications is found to be limited in various fields owing to their high toxicity at their optimum dose levels. Therefore, a worldwide hunt is on to find an ideal radio-protective agent for its uses against planned and unplanned radiation exposure. For this purpose Swiss albino mice were divided in various groups. Group I was sham irradiated and served as normal. Group II was given mercuric chloride solution at the dose of 0.5ppm. Group third was exposed to 2.5 Gy of gamma radiations; Group IV was treated with gamma radiation and mercuric chloride. Group V, VI and VII were given *Moringa oleifera* seven days prior to radiation or mercuric chloride or combined treatment. The animals from all experimental groups were sacrificed by cervical dislocation at each post-treatment interval of 1,2,4,7, 14 and 28 days. After sacrificing the animals, pieces of the liver were taken out and kept at -20°C for various biochemical parameters. The value of total proteins, glycogen, acid phosphatase & alkaline phosphatase activities and RNA increased whereas the values of cholesterol and DNA declined. Almost normal values were noticed on day-28. After combined treatment of radiation and mercuric chloride the changes were more severe showing synergistic effects. An early and fast recovery in the drug treated groups showing protection provided by the drug.
Banot et al. (2017) investigated the impact of the *Moringa oleifera* against combined administration of mercuric chloride and radiation on histopathological changes in the liver of mice. The mice were exposed to mercuric chloride (0.5 ppm) and gamma radiation (5.0 Gy) simultaneously and individually. The experimental groups were given *Moringa oleifera* seven days prior to radiation or mercuric chloride treatment. The changes included cytoplasmic degranulation, vacuolation, nuclear pycnosis, necrosis, hyperaemia and leucocytic infiltration etc. In the combined treatment groups the changes were more severe showing synergistic effect. An early and fast recovery in *Moringa* pre treated groups may be due to the protection provided by the drug.