The present study was an endeavor to determine the ameliorative potential of aqueous *Aloe vera* gel extract against deleterious effects induced by whole body X-ray exposure on various organs in balb/c mice.

- Phytochemical analysis of various constituents in aqueous *Aloe vera* gel extract indicated the presence of carbohydrates, proteins, anthraquinones tannins and flavonoids. The IC50 value in the extract by ABTS and DPPH assay was found to be 20 µg/µl and 50 µg/µl respectively depicting its antioxidant property. Sodium, potassium, calcium, magnesium, iron and zinc were found in abundance in the extract as indicated by WD-XRF technique.
- To select the safe dose of aqueous *Aloe vera* gel extract, male balb/c mice were randomly divided into four groups: Group I served as control. Group II, Group III and Group IV animals were p. o. administered with *Aloe vera* extract at a daily dose of 250, 100, 50 mg/ kg b.w. respectively for 30 days.
- Liver of control animals revealed normal histoarchitecture comprising of hepatocytes dorsally radiating from the central vein to portal area forming the hepatic cords. However, animals fed with higher doses of *Aloe vera* gel extract (250 mg/ kg b.w. and 100 mg/ kg b.w.) revealed cell degeneration, enlargement of central vein with congested sinusoids, while lower dose of *Aloe vera* extract (50 mg/ kg b.w.) exhibited normal hepatic histoarchitecture.

- Spleen from control animals exhibited normal tissue architecture consisting of white pulp with intact germinal center surrounding lighter marginal zone and red pulp. *Aloe vera* administration (250 mg/ kg b.w. and 100 mg/ kg b.w.) revealed widespread infiltration in white pulp, decreased white pulp, increased red pulp, dilated sinusoids and animals fed with 50 mg/ kg b.w. of *Aloe vera* gel extract showed normal histoarchitecture with germinal center surrounding the marginal zone separating the red pulp from the white pulp area.
- Histopathological examination of kidney from control animals revealed normal architecture with regular glomeruli in the cortex and cortico-medullary region. Animals administered with *Aloe vera* gel extract (250 mg/ kg b.w. and 100 mg/ kg b.w.) resulted in atrophy of renal corpuscle, decrease in glomerular cellularity, reduction in number of Bowman's capsule, glomerular congestion with increased Bowman's spaces, glomeruli infiltration in cortex region and crowding of nuclei with hyperplastic response in inner medulla. Administration of *Aloe vera* gel extract at the dose of 50 mg/ kg b.w. showed normal cortical labyrinth and medullary region.
- Testicular section of control animals showed normal histoarchitecture which consists of well-organized seminiferous tubules with complete spermatogenesis and normal interstitial connective tissue. Administration of *Aloe vera* gel extract to animals (250 mg/ kg b.w. and 100 mg/ kg b.w.) resulted shrunken tubules, disorganized/ distorted seminiferous tubules,

depletion in cell population, cellular inflammation, abnormal widening of interstitial spaces, while animals treated with 50 mg/ kg b.w. of *Aloe vera* gel extract revealed normal histoarchitecture of testes.

- Elevation in the activity of LDH (tissue injury marker) was observed in serum and various tissues upon administration of higher doses of *Aloe vera* gel extract (250 mg/ kg b.w. and 100 mg/ kg b.w.). However, LDH activity remained unaltered on administration of 50 mg/ kg b.w. *Aloe vera* gel extract.
- Increased LPO level in blood and various tissues was observed upon administration of *Aloe vera* gel extract (250 and 100 mg/ kg b.w.) when compared to control group. However, it remained unaltered upon administration of *Aloe vera* gel extract at the dose of 50 mg/ kg b.w.
- For the standardization of whole body X-ray exposure, male balb/c mice were randomly divided into two groups: Group I mice served as control. Group II (a) mice were exposed to X-ray (twice a day) for two consecutive days (27^{th} and 28^{th}) in the last week of the study. Group II (b) mice were exposed to X-ray (twice a day) for four consecutive days (27^{th}, 28^{th}, 29^{th} and 30^{th}) in the last week of the study. The present study showed 2 fold elevation in LPO levels and LDH activity after two days of X-ray exposure. However, significant increase (3 folds) in the activity of LDH, levels of LPO and histopathological alterations were observed after four days of X-ray exposure (2Gy) when compared to two days of exposure (1Gy) in blood and various tissues of mice. Therefore, 2Gy of X-ray irradiation was selected as near to the lethal dose for further investigations.

- Histopathological analysis of liver from control animals revealed normal histoarchitecture. Liver of X-ray irradiated mice showed in widening and dilated central vein associated with ruptured endothelial lining cells with congested plates of vascular sinusoidal spaces along with vascular channels radiating out from central veins when compared to control group.
- Spleen from control animals revealed normal histoarchitecture. X-ray irradiated animals showed decrease in the amount of white pulp, excessive amount of red pulp, large number of macrophages and lymphocytes number in red pulp. Infiltration in white pulp and thickened trabeculae were also observed.
- Cortical and medullary section of kidney from control group revealed normal histoarchitecture. Kidney of X-ray irradiated animals exhibited glomerular attenuation and glomerular congestion, reduction and shrinkage of Bowman's capsule. The thick and thin parts of loop of Henle and collecting ducts present in inner medulla revealed normal histoarchitecture when compared to control group.
- Histopathological analysis of testes from control animals revealed normal architecture. Testes of X-ray irradiated animal revealed shrinkage of seminiferous tubules, empty tubules, derangement of cellular organization due to germ cell degeneration and disrupted basement membrane. Lumen was full of cellular and spermatogenic debris and thinning of seminiferous epithelium with loosely arranged cells were also observed as compared to control animals.

- After these standardizations, male balb/c mice were divided into four groups. Group I [control]. Group II [administration of aqueous *Aloe vera* gel extract (50 mg/ kg b.w.) on alternate days for 30 days]. Group III [X-ray (twice a day) for four consecutive days in the last week of the study] Group IV [aqueous *Aloe vera* gel extract administration followed by X-ray irradiation as explained previous].
- Control and *Aloe vera* treated animals did not depict any alterations in the body weight diet intake and water consumption during the entire course of treatment. X-ray irradiated animals exhibited a significant decline in the body weight and diet intake on last week of the study. Likewise, the body weight and diet intake was comparatively increased in *Aloe vera* + X-ray irradiated group in 4^{th} week when compared to only X-ray irradiated group. Moreover, water consumption remained unaltered in all the treatment groups until the end of the treatment period.
- Histopathological analysis of liver from control animals revealed normal histoarchitecture. Histological alterations such as widening and dilated central vein and ruptured endothelial lining cells with congested plates of vascular sinusoidal spaces were observed in liver of X-ray irradiated animals when compared to control animals. However, *Aloe vera* + X-ray irradiated mice revealed normal histoarchitecture of liver.
- Histopathological examination of spleen from control animals revealed normal architecture with normal areas of white pulp with germinal center and surrounding lighter marginal zone and red pulp. Spleen of X-ray

irradiated animals showed decreased amount of white pulp, increased red pulp, large number of macrophages, reduction in a number of lymphocytes in red pulp, thickening of trabeculae and infiltration in white pulp. *Aloe vera* + X-ray irradiated group indicated decreased white pulp and considerably less macrophages when compared to X-ray irradiated group.

- Histopathological examination of kidney from normal animal revealed normal cortical labyrinth and medullary region. X-ray irradiated animals revealed decrease in number of Bowman's capsule, shrinkage of Bowman's capsule, glomerular attenuation and glomerular congestion. Moreover, inner medullary section revealed normal histoarchitecture. Regarding *Aloe vera* + X-ray irradiated mice resulted in normal histoarchitecture of cortex and medulla as observed in control group.
- Testes of control animals showed normal histological features in seminiferous tubules. By contrast, marked alterations in the histoarchitecture of testes were observed in X-ray irradiated mice revealed depleted germinal cell population, disrupted basement membrane, empty tubules, shrunken and disorganized/ distorted seminiferous tubules. Lumen was full of spermatogenic debris and thinning of seminiferous epithelium with loosely arranged cells was also observed when compared to control animals. *Aloe vera* + X-ray irradiated animals resulted in organized histoarchitecture of seminiferous tubules when compared to X-ray irradiated group.
- Whole body exposure to X-ray exhibit a significant increase in LDH activity of serum, liver, kidney and testes when compared with control and *Aloe vera*

treated groups. *Aloe vera* + X-ray irradiated mice caused a significant decrease in LDH activities when compared with X-ray irradiated mice.

- The liver enzymes (SGPT, SGOT, T-BIL, D-BIL, A/G ratio) were significantly elevated by X-ray irradiation when compared to control and *Aloe vera* treated groups. *Aloe vera* + X-ray irradiated group showed significantly decreased activity when compared to X-ray irradiated group.
- Urea, creatinine and blood urea nitrogen levels were significantly increased in X-ray irradiated group. *Aloe vera* + X-ray irradiated group showed significantly decreased activity when compared to X-ray irradiated group. A significant decrease was also observed in GFR level of X-ray irradiated animals. However, *Aloe vera* + X-ray irradiated group showed significantly elevation in GFR level when compared to X-ray irradiated group.
- X-ray irradiation caused significant reduction in sperm motility sperm count as well as testosterone levels as compared to control and *Aloe vera* group. *Aloe vera* + X-ray irradiated groups showed significant elevation in sperm count, sperm motility and testosterone level when compared to X-ray irradiated group.
- A significant increase was observed in percentage of micronucleus in splenic tissue of X-ray irradiated group indicating abnormal, uncontrolled and unequal cell division. *Aloe vera* + X-ray irradiated mice caused a significant decrease in micronucleus formation when compared with X-ray irradiated group demonstrated the protective effects of *Aloe vera* extract.

- Moreover, after X-ray exposure a significant increase in the percentage incidence of aberrant cells in hepatic tissue was observed when compared with control and *Aloe vera* treated group. X-ray irradiation caused pulverization, breaks, deletions and multiple types of abnormal rearrangements in the chromosomes. However, extensive pulverization was observed in X-ray irradiated group and mild to moderated pulverization was observed in *Aloe vera* + X-ray irradiated group. *Aloe vera* + X-ray irradiation significantly reduce the increase in percentage incidence of aberrant cells and abnormalities when compared to X-ray irradiated group. Reduction in micronuclei formation and chromosomal aberrant cells in *Aloe vera* pretreated and irradiated group indicated the amelioration of X-ray induced cellular damage after *Aloe vera* administration.
- TUNEL assay in liver, spleen, kidney and testes in X-ray irradiated group showed higher apoptotic cells when compared to control. However, *Aloe vera* + X-ray irradiated group revealed lesser amount of apoptotic cells as compared to X-ray irradiated animals.
- Genomic DNA of X-ray irradiated group showed a diffused and mild smear indicating DNA damage in different tissues. An intact genomic DNA band was observed in control and *Aloe vera* treated group indicating the integrity of DNA.
- Increased ROS and LPO levels were observed in the X-ray exposed group when compared to control group. However, decrease in ROS and LPO levels was observed in the animals of *Aloe vera* + X-ray exposed group.

- Reduction in GSH content and activities of GSH-based antioxidant enzymes i.e. GSH-Px and GR were found to be increased in X-ray irradiated group when compared to control group in plasma and various organs. Also, an enhanced activity of SOD in plasma, kidney and testes and activity was found to be decreased in liver and spleen tissues of X-ray irradiated animals. GST and CAT activity in plasma and various organs was found to be increased in X-ray exposed group. However, increased SOD activity in liver and spleen, while reduction in plasma, kidney and testes were observed in *Aloe vera* + X-ray exposed animals when compared to X-ray irradiated animals. Also, reduction GR, GSH-Px, GST, CAT activity was observed in *Aloe vera* + X-ray irradiated group when compared to the animals that are exposed to X-ray only.
- X-ray exposure led to a significant enhancement in neutrophil counts when compared to control and *Aloe vera* treated mice. In contrast, *Aloe vera* + X-ray exposed mice induced a marked decrease in neutrophil counts in comparison to X-ray exposed group. TLC was found to be enhanced in X-ray irradiated group. However, *Aloe vera* + X-ray irradiated group showed decrease in TLC and neutrophils in comparison to control mice. Moreover, platelet counts were significantly reduced by X-ray irradiation when compared to control group and *Aloe vera* treated groups. *Aloe vera* + X-ray irradiated group showed significant reduction in platelet counts when compared to X-ray irradiated group.

- In addition, inflammatory markers (TNF-α, IL-6) were significantly elevated by X-ray irradiation when compared to control group and *Aloe vera* treated groups. *Aloe vera* + X-ray exposed group showed significantly decreased levels of TNF-α and IL-6 when compared to X-ray irradiated group.

Conclusion

In conclusion, Aloe vera pretreatment reduced radiation- induced damage by protecting against the radiation-induced histopathological, biochemical, hematological and DNA damage. Free radical scavenging, elevation in antioxidant status, reduction in lipid peroxidation and DNA damage appear to be important in providing radioprotection due to its antioxidant, anti-apoptotic and anti-inflammatory potentials.