



Effect of *Opuntia elatior* on Alteration in Glutamic Oxaloacetic Transaminase activity induced by Gamma Radiation in Swiss Albino Mice

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Abstract : When mice are exposed to gamma radiation, all the body systems are affected adversely. Liver enzymes which are responsible for removal of toxicity created by irradiation increases in the body. Several herbs are known to reduce this toxicity. *Opuntia elatior* (OE), a cactus, was tested in this study for its activity to remove toxins induced by whole body exposure to Co⁶⁰ gamma rays (6Gy). A moderate dose of OE (10 mg/kg body weight) was given orally to mice, before whole body irradiation and GOT activity was studied in their kidney at 1, 3, 5, 10, 15 and 30 post treatment days. Whole body gamma ray exposure increases GOT activity in the kidneys also. In OE pretreated animals this increase was significantly lesser and early recovery was observed. Two types of extracts were used in this study (ethanolic and acetonic). Both were able to reduce gamma radiation induced increase significantly and almost equally. As OE cladode extracts are quite cheap and produce no side effects (OE cladodes are used as food), it may be used to prevent unwanted toxic effects of gamma radiation exposure.

Key words: Gamma radiation, Glutamic Oxaloacetic Transaminase Activity, Mouse kidney, *Opuntia elatior* extract.

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1. INTRODUCTION

Radiation exposure of human beings is either accidental such as during nuclear explosions, war or terrorist activity or planned as used for medical purposes. Body respond to it and suffers the damage. Cellular radiosensitivity is directly related to dividing capacity of the body cells and gastrointestinal, hematopoietic, gonadal and skin tissues of mammals comparatively suffer more due to irradiation.^{1,2,3,4,5} Kidneys are moderately radiosensitive. Minor but important structural and functional changes are known. Rats treated with gamma rays showed atrophied glomeruli, proximal and distal convoluted tubules had nuclear alterations with pyknosis and karyolysis.⁶ *Opuntia elatior*, a food item in some parts of the world, is known to have several neutraceuticals and pharmacologically active compounds.^{7,8,9} Different parts of OE (*Opuntia elatior*) contains high levels of betalain, taurine, calcium, magnesium and antioxidants. Most of the radioprotective plant products are free radical scavengers, antioxidants and inhibits lipid peroxidation. These extracts not only reduces detrimental effects of ionizing radiation exposure but also known to increase survival rates. Most of these compounds are aromatic. *Opuntia* sp. are distributed world wide and grow in xerophytic conditions without much efforts. Thus their product will be cheap and easily available. With the increasing use of radiations in various fields of life, chances to be exposed to them are increasing and need to develop cost effective radioprotectors without side effects is increasing. Amifostine, the only clinically used radioprotector is very expensive and has highly toxic side effects. Hence development of an effective and cheap radioprotector from *Opuntia elatior* may prove be useful in handling gamma radiation induced adverse effects. In our study, *Opuntia elatior* (OE) was tested for its activity to remove toxins induced by whole body exposure to Co⁶⁰ gamma rays (6Gy).

2. MATERIALS AND METHODS

Male Swiss albino mice (*Mus musculus norvegicus*) 6-8 weeks old, 25±2 g each from inbred colony were selected for the experiments. They were maintained under controlled conditions of temperature 37±5° C and kept in natural day light and darknight cycles. Animals were irradiated with Gamma radiation at Cobalt teletherapy unit (ATC-C9 at Cancer Treatment Center, Radiotherapy Department, SMS Medical College and Hospital, Jaipur, Rajasthan, India. *Opuntia elatior* cladode cut into pieces, dried and then powdered. The extract of the stem of *Opuntia elatior* was made in Ethanol at 68 °C and Acetone at 40°C in a Soxhlet apparatus for 36 hours each. The optimum dose was selected on the basis of survival experiments. Selected dose of plant extract was dissolved in double distilled water (DDW). The animals were fed one extract at a time by gastric intubation with at the rate of 10mg/kg body weight, according to the plan of experiments.

2.1 Ethical committee

The experiment was approved by Ethical committee of the Department of Zoology, University of Rajasthan, and Jaipur. (CPCSEA registration no. 1678/Go/Re/S/12/ CPCSEA dated 16.06.2017.)

2.2 Design of experiment

Adult, healthy, Swiss albino mice were used for the study.

They were divided into four groups.

- Group I: Control mice without any treatment.
- Group II: 6 Gy Co⁶⁰ gamma radiation only.
- Group III: *Opuntia elatior* extract (10mg/kg body weight) only.
- Group IV: *Opuntia elatior* extract (10mg/kg body weight) + 6 Gy Co⁶⁰ gamma radiation

Then mice were sacrificed on autopsy intervals as 1, 3, 5, 10, 15, 30 day. Kidney was removed for analysis of Glutamic oxaloacetic activity by Thefeld, wallnofer, bergmeyer method.^{10,11,12}

3. STATISTICAL ANALYSIS

The results obtained were expressed as mean±standard error (S.E.). The statistical difference between various groups analyzed by two way ANOVA for comparing the results obtained. Data was analyzed by two way anova followed by tukey's post hoc test. n=6, ***P<0.001 (Highly significant), **P<0.01 (Moderate significant), (Less significant) *P<0.05, Without superscript*, non significant

4. RESULTS

In the present experiment, it was observed that in acetone extract treated mice the GOT/AST activity in the control group (without any treatment) was almost constant from day 1st to 30th. In the group treated with only plant extract, the activity decreased till day 10th but improved by day 30th post treatment (53.24 to 58.78 mg/gram). In the Group treated with 6 Gy radiations only, it was observed that the activity significantly increased at day 30th after treatment (64.84 to 81.32 mg/gram). In the group treated with radiation along with plant extract, the activity decreased insignificantly by day 30th post treatment (65 to 55.81 mg/gram) which is almost equal to the normal activity of the control group. (Table 1, Figure 1) GOT/AST activity, in the control group mice (without any treatment) was observed, almost constant from day 1st to 30th. Further in the group treated with only ethanolic plant extract, the activity significantly decreased by day 30th post treatment (50.08 to 47.48 mg/gram). In the group treated with 6 Gy radiation only, it was observed that the activity significantly increased by day 30th post treatment from day 1 (71.47 to 82.33 mg/gram). In the group treated with radiation along with ethanolic plant extract, it was observed that the activity decreased insignificantly by day 30th post treatment (67.41 to 55.60 mg/gram) which is almost equal to the activity of the control group. (Table 2, Figure 2)

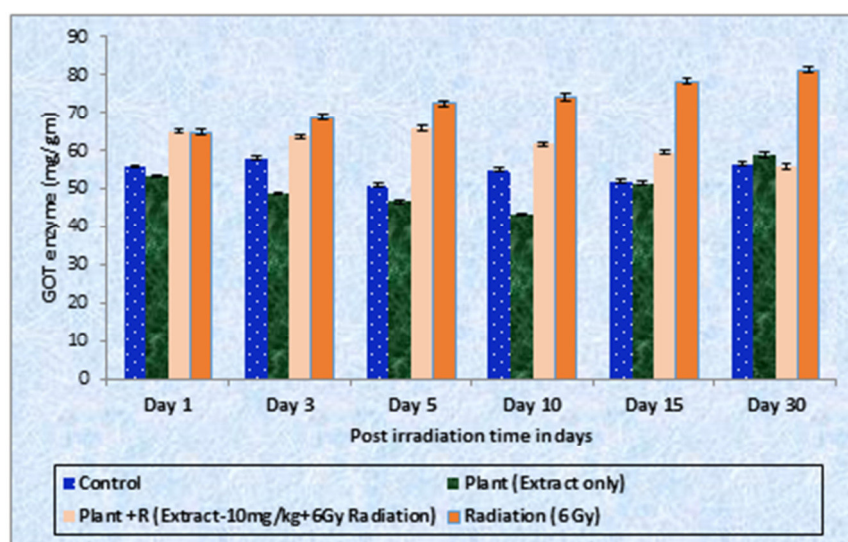
4.1 Two way anova (Two factor, 1. Treatment 2. Day)

When anova test was applied, it was observed that in *Opuntia elatior* acetone extract treated mice along with day, without day, individually day activities of GOT were less significant. In *Opuntia elatior* ethanol extract treated individually day was less significant and individual treatment was moderately significant and treatment along with day was highly significant. (Table 3, 4) From the observations, concluded that both the extracts of *Opuntia elatior* cladode, Acetone and Ethanol, almost equally recovered the GOT activity against gamma radiation, which is statistically significant?

Table 1. Variation in GOT/AST activity of irradiated mouse with and without *Opuntia elatior* extract (Acetone) treatment

Autopsy interval Experimental Groups	Day1	Day3	Day 5	Day10	Day 15	Day 30
Group 1 Control (without any treatment)	55.81±0.48	57.82±0.52	51.06±0.55	54.86±0.47	52.09±0.44	56.54±0.48
Group 2 (Plant Extract only)	53.24±0.45	48.81±0.41	46.53±0.39	43.24±0.38	51.35±0.46	58.73±0.58
Group 3 (Plant Extract-10mg/kg+6Gy irradiation)	65.00±0.66	63.61±0.59	65.98±0.66	61.72±0.57	59.52±0.52	55.81±0.52
Group 4 Radiation (6 Gy)	64.82±0.64	68.72±0.70*	72.46±0.77*	74.10±0.77*	78.30±0.80*	81.32±0.84*

Data represented by Mean±SE (Six mice per group).n=6,P<0.05(Data with superscript* were significant and data without superscript* were on significant).Data represent the mean value at that autopsy interval.

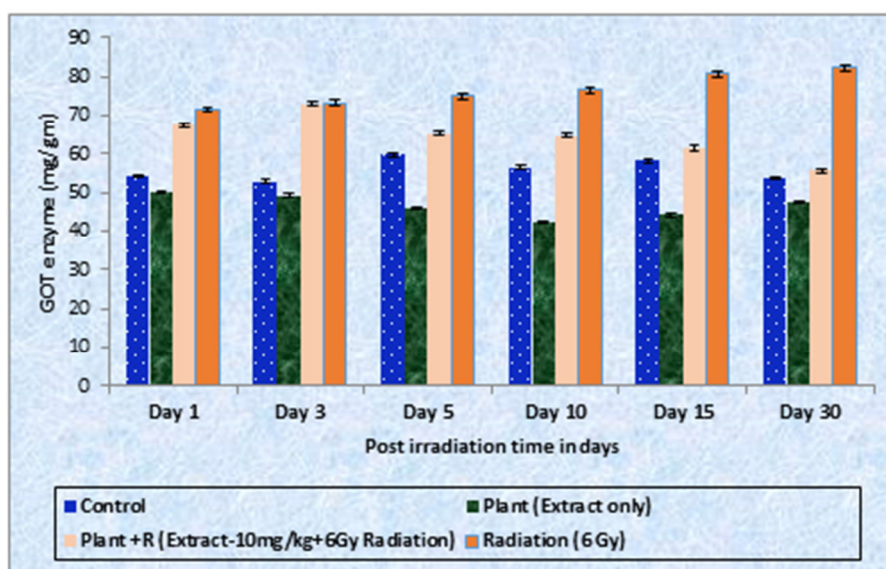


Data represented by Mean±SE (Six mice per group).n=6,P<0.05(Data with superscript* were significant and data without superscript* Were non significant).Data represent the mean value at that autopsy interval.

Fig 1. Variation in GOT/AST activity of irradiated mouse with and without *Opuntia elatior* extract (Acetone) treatment**Table 2. Variation in GOT/AST activity of irradiated mouse, with and without *Opuntia elatior* extract (Ethanol) treatment**

Autopsy interval Experimental Groups	Day1	Day3	Day 5	Day10	Day 15	Day 30
Group 1 Control (without any treatment)	54.20±0.47	52.87±0.46	59.75±0.54	56.54±0.49	58.17±0.53	53.68±0.44
Group 2 (Plant Extract only)	50.08±0.44	49.22±0.43	45.97±0.45	42.21±0.40	44.2±0.44	47.48±0.46
Group 3 (Plant Extract-10mg/kg+6Gy irradiation)	67.41±0.61	73.02±0.68	65.26±0.64	64.67±0.62	61.52±0.60	55.60±0.53
Group 4 Radiation (6 Gy)	71.47±0.69*	73.33±0.71*	75.15±0.77*	76.58±0.76*	80.75±0.81*	82.33±0.84*

Data represented by Mean±SE (Six mice per group).n=6,P<0.05(Data with superscript* were significant and data without superscript* Were non significant).Data represent the mean value at that autopsy interval.



Data represented by Mean \pm SE (Six mice per group).n=6,P<0.05(Data with superscript* were significant and data without superscript* werenon significant).Data represent the mean value at that autopsy interval.

Fig 2. Variation in GOT/AST activity of irradiated mouse, with and without *Opuntia elatior* extract (Ethanol) treatment

Table 3. Variation in GOT/AST activity of irradiated mouse with and without *Opuntia elatior* extract (Acetone) treatment. Two way anova (Two factor, 1.Treatment 2.Day)

Two way anova factor	Df	Sum Sq	Mean Sq	F Value	Significance
Treatment	5	1212.36	588.69	169.38	*
Day	4	8.64	5.99	5.92	*
Treatment: Day	20	362.31	41.48	28.664	*

Data was analyzed by two ways anova followed by tukey's post hoc test. n=6,***P<0.001(Highly significant),**P<0.01(Moderate significant), (Less significant)*P<0.05,Without superscript*,non significant

Table 4. Variation in GOT/AST activity of irradiated mice with and without *Opuntia elatior* extract (Ethanol) treatment. Two way anova (Two factor, 1.Treatment 2.Day)

Two way anova factor	Df	Sum Sq	Mean Sq	F Value	Significance
Treatment	5	1134.79	564.81	151.17	**
Day	4	5.41	5.56	5.69	*
Treatment : Day	20	337.89	36.95	22.644	***

Data was analyzed by two ways anova followed by tukey's post hoc test. n=6,***P<0.001(Highly significant),**P<0.01(Moderate significant), (Less significant)*P<0.05,Without superscript*,non significant

5. DISCUSSION

Increased GOT activity in irradiated animals is due to radiation toxicity which is clearly visible in the kidney of mice. Pradeep et al, reported that marked elevation in the activities of serum AST/ GOT, ALT, ALP, and LDH after exposure of rats to γ -radiation at doses of 1, 3 and 5 Gy.¹³ It is actually because of secretion of those enzymes from the cytoplasm into blood circulation at rapid rate after rupture of the plasma membrane and cellular disruption¹⁴. Marzook et al, reported radioprotective effect by *Costus speciosus* treatment.¹⁵ There are plenty of reports on radioprotective effect by plant product treatment.⁹ Shivabasavaiah and Krishna Ram¹⁶, reported ethnomedicinal value of *Opuntia elatior* fruits. The fruit extract was tested for presence of various bioactive compounds, which contain alkaloids, carbohydrates, oils, flavonoids, phenolics, fats, tannins, steroids, and saponins. The oral administration of crude

extract exhibited no noxious effect on the external morphology and body weight of the mice. Thus, it established a scientific base for further use of *Opuntia elatior* fruit for pharmacological tests such as antibacterial, antidiarrheal, anti-inflammatory, antimicrobial, analgesic and antidiabetic properties. Patil et al, reported that *Opuntia ficus indica* has antioxidant activity because of beta alanins, phenols, vitamin C and beta carotene.¹⁷ On the other hand study conducted on mice kidney histology to record radiation pathology, there was little change after 6 Gy gamma irradiation. On 1st day congested glomeruli, degenerated proximal convoluted tubules and distal convoluted tubules were observed with pycnotic nuclei and necrosis in a few cells. Recovery was observed in Acetone and ethanol extract of OE treated mice at the rate of 10 mg/kg body weight. Both extracts of OE cladode protect histology of kidney in whole body gamma irradiated animals. Arsad et al, found *Rhaphidiphora decurva* (Roxb) effective in reducing histopathological alterations in

kidney.¹⁸ *Annona muricata* leaf extract also protects kidney's histology. *Averrhoa carambola* leaf extract is also reported to protect kidney against irradiation^{19,20}. In the present study phytoconstituents and vitamins present in the OE cladode extract might have protected unwanted increase in GOT activity in mouse kidney. Poonia and Sharma, have already reported radioprotection by OE of mouse blood after total body irradiation.²¹ They observed that increase in blood glucose level, cholesterol level (HDL, LDL, VLDL and total cholesterol) were also protected by the same type and dose of OE extract.^{22,23,24}

6. CONCLUSION

In this research work, GOT/AST (Glutamate oxaloacetic transaminase/Aspartate aminotransaminase) enzyme activity was effected by gamma radiation in Swiss albino mice. Results showed that after post-treatment by gamma radiation at dose rate 6 Gy, GOT/AST activity in Swiss albino mice was increased and after *Opuntia elatior* cladode extract, GOT/AST activity was restored to normal. *Opuntia elatior* cladode extract prepared in ethanol and acetone and fed by gastric intubation to the mice at the dose rate of 10 mg/kg body

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weight and both the extracts were found to be almost equally potent to prevent, the alteration in GOT/AST activity by gamma irradiation.

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8. AUTHORS CONTRIBUTION STATEMENT

Kavita Poonia and Professor Jaimala Sharma both contributed to the research article by means of experimental work and wrote the research article.

9. CONFLICTS OF INTEREST

Conflicts of interest declared none.

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