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Research Article

EFFECT OF *OPUNTIA ELATIOR* ON ALTERATION IN BLOOD GLUCOSE LEVEL INDUCED BY GAMMA RADIATION IN MICE

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ABSTRACT

The modulatory effect of *Opuntia elatior* stem extract was studied in Swiss albino mice at various two doses i.e. 6 Gy and 8 Gy of gamma radiation in the presence (experimental) or absence (control) of *Opuntia elatior* extract (10 mg/kg body wt.) to observe in blood glucose level alterations. These animals were scarified and their blood was collected at days 1, 15 and 30 post-irradiation. The glucose level was scored almost constant in the control group(without any treatment) and increased in gamma radiation treated mice group, whereas a recovery pattern was recorded in experimental animals (in only plant extract treated and plant extract with radiation treated mice's groups and a normal value was achieved by at 30th day after dose level.

Key words:- *Opuntia elatior*; Gamma radiation; glucose level; Mice.

INTRODUCTION

Radiations are divided into two classes on the basis of their energy potential, ionizing and non-ionizing radiation. Alpha, beta, gamma and X rays have enough energy to cause ionization when interact with matter thus named as ionizing radiation. Occupational or accidental exposure to radiation via nuclear weapons in war and terrorist activities and also the use of radioactive materials in electricity generation are the major concern to the increasing risk of radiation exposure [1,2]. The biological concerns of after contact with ionizing radiations are interceded by a series of physical, chemical, biochemical and cellular responses originated after the deposition of the radiation in the medium [3]. The energy related with ionizing radiation is significantly greater than the bond energies of many molecules and can cause hemolytic bond scission and the generation of secondary electrons.

In recent scenario attention has been devoted worldwide to screen for non-toxic radioprotectors that can be recommended as defense against disastrous effects of radiation in livelihood along with salutary situations for human beings. Though there are lot of reports and till date no molecule has been innovated as an effective non-toxic radio protector for their use[4,5,6]. Therefore, screening of bioactive compounds represents a major source for the discovery of new radio protective drugs.

Administration of glucose level not having any harmful effect is still a big task to the various researchers working in area of medicinal biology. The consumption of the drugs is limited by their pharmacokinetic stuffs, including failure rates, and associated toxic effects [7]. In spite of remarkable innovations in the treatment of increased glucose level by oral hypoglycemic agents, keen innovations for fresher drugs is still in progress as current synthetic drugs have many drawbacks[8]. The existing treatments for control of glucose level have insulin and oral antidiabetic mediators like biguanides, sulfonylurea and α-glycosidase inhibitors. Most of these drugs have serious side effects. Thus, the administration of diabetes without toxic effects is still a tedious task[9]. So, keen interest in bioactive compounds because of their effectiveness, least toxicity in clinical experience, and relatively low costs has attracted lot of researchers.

Opuntia is a great genus of succulent shrubs, native of the new world, now broadly cultivated in the parts of the world which has higher temperatures, due to their exceptional morphology and gorgeous flowers. Generally they are known as called as Prickly pears, as they have edible fruits. These are initially introduced into India and other eastern countries by early European communities. The fruit of plant is has important role as refrigerant, and used in treatment in gonorrhea. In addition to food, Indian fig is used to combat with whooping cough, diabetes, prostate problems, rheumatism, nosebleed, and in dentistry in central Mexico [10]. In Australia it is consumed to lower the blood sugar level in diabetes. The baked fruit is used in whooping cough and syrup of the fruit helps in secretion of bile and combat with expectoration and spasmodic cough[11].The flowers cure bronchitis and asthma. A tea is prepared from flowers used in combating kidney problems. Dried flowers are also ground into a paste and applied to the skin for measles[12,13].

MATERIALS AND METHODS

Male *Swiss albino mice* (*mus musculus norvegicus*) 6-8 weeks old, weighing 25±2 g each from an inbred colony at Department of Zoology, University of Rajasthan, Jaipur, were selected for the experiments. They were maintained under controlled conditions of temperature 37±5° C and kept in natural day light and dark night cycles.

Institutional animal ethics committee (IAEC) approval number of Department of Zoology, University of Rajasthan, Jaipur, is CPCSEA registration no. 1678/Go/Re/S/12/CPCSEA dated 16.6.2017.

Source of Irradiation

Cobalt teletherapy unit (ATC-C9) was used to irradiate the animals at Cancer Treatment Center, Radiotherapy Department, SMS Medical College and Hospital, Jaipur, Rajasthan, India. Animals were controlled in well ventilated perspex boxes (30cm×30cm× 5cm) and whole body was exposed to gamma radiation with external Co^{60} teletherapy, with source surface distance (SSD) of 80cms to transport the dose level of 1.47Gy/min. The dose rate was standardized during the whole research work following the decay table of Co^{60}.

Source of plant material

The plants were harvested from Smriti van, Jaipur, Rajasthan and voucher specimen was deposited in Department of Botany, University of Rajasthan for authentication (RUBL no. 211574).

Preparation of Plant Extract

Opuntia elatior stem was peeled off, cut into pieces and shade dried and then powdered. Plant material was taken out and shade dried and then powdered. The extract of stem of *Opuntia elatior* was prepared by extracting powder in ethanol and acetone separately at 68ºC and 40ºC in a Soxhlet apparatus for 36 hours each. The extract was lyophilized in double distilled water (DDW). The animals were fed one extract at a time by gastric intubation, according to the plan of experiments. Both ethanolic and acetonic extracts were prepared at 10mg/kg body weight. (On the basis of survival assay).

Experimental design

Survival assay for dose selection

The animals were witnessed sensibly daily for symptoms of radiation sickness, behavioral toxicity and mortality. The groups which were irradiated without plant dose had symptoms of radiation sickness within 2-4 days after exposure to 8Gy gamma radiation. Mice for present research were divided into 5 different groups of 5 animals each and were given *Opuntia elatior* extract dissolved in DDW (Double distilled water) through oral gavages as 5, 10, 20, 40, 80 (mg/kg body weight/day) for 5 consecutive days. After 30 minutes of the last administration of *Opuntia elatior* extract (i.e. on day 5th) animal were exposed whole body to 8 Gy gamma radiation. Animals for optimum dose selection were observed till 30 days, after radiation exposure. The optimum dose was chosen as 10 mg/kg body weight/day for both *Opuntia elatior* extract in ethanol and acetone, on the basis of radiation sickness and survival of mice. Ethanol and acetone survival assay curve for optimum dose selection of *Opuntia elatior* extract is figure 1 and figure 2.

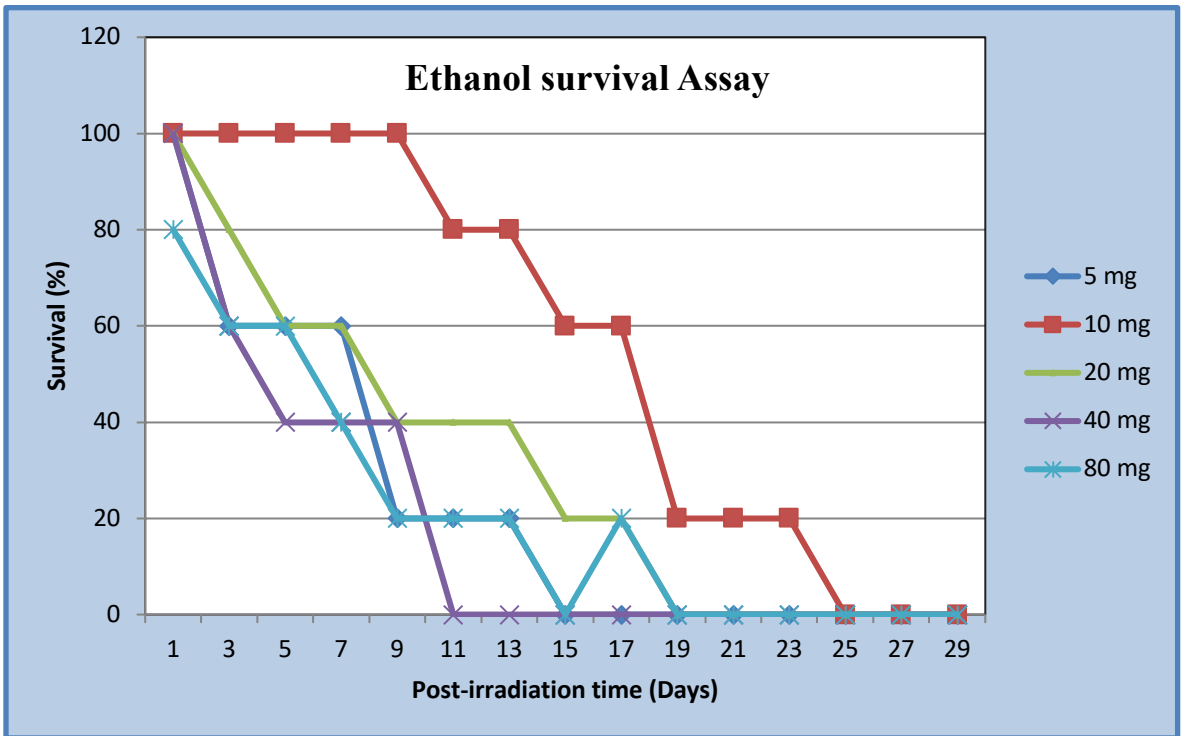

Figure 1. Animals for optimum dose selection of *Opuntia elatior* extract in Ethanol were observed till 30 days after radiation exposure.

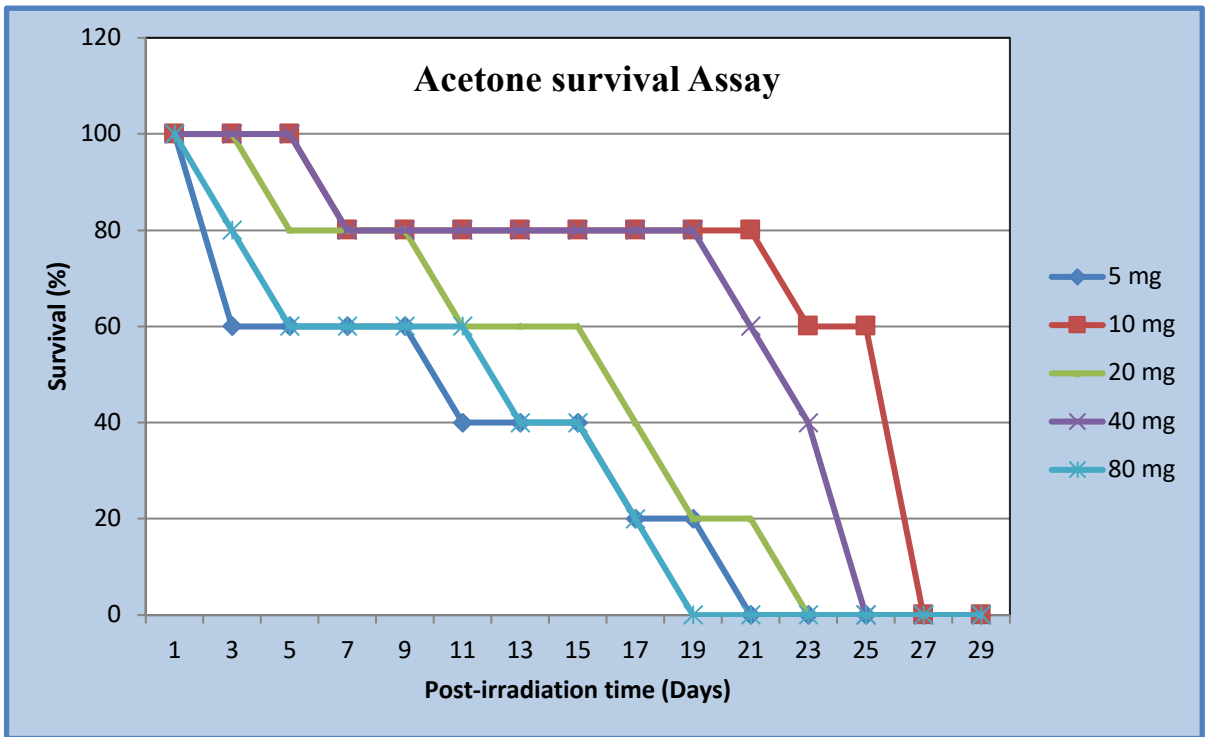

Figure 2. Animals for optimum dose selection of *Opuntia elatior* extract in acetone were observed till 30 days after radiation exposure.

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 Co 60gamma radiations 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

The animals were observed for changes in their behavior, body weight, mortality (if, any). The animals will be sacrificed at suitable post treatment intervals.

Autopsy

The animals from all the groups were sacrificed by cervical dislocation at 1, 15 and 30 days after irradiation. Six animals were sacrificed at each interval from every group and hematological parameters were studied.

Glucose

Serum glucose was assayed on the basis of Autospan glucose kit based on GOD-POD end point assay (Kaplan & Lavernel 1983)

Calculation

Serum /Plasma Glucose ($mgdL^{-1}$) = Absorbance of test x conc. of standard / Absorbance of standard

Statistical analysis

The result for all the groups at various necropsy intervals were expressed as mean ± Standard error (S.E.). To find out whether mean of sample drawn from experimental (group IV) deviates significantly from respective control (group I), for this Two way ANOVA was applied.

RESULTS

***Opuntia elatior* extract in Ethanol**

In Control group (without any treatment) the level of glucose is almost constant at all autopsy intervals (1^{st},15^{th},30^{th}).When only plant extracts were administrated there was decrease in glucose level, which was found to be maximum at day 1^{st} and reduced thereafter (89 mg/dl)at day 30^{th}.When only radiation (6 Gy) given to mice ,the glucose levels were increased at all autopsy intervals 1^{st},15^{th},30^{th}day,as compared to the normal glucose level(Control group).When plant extracts were administrated to mice irradiated with gamma radiation there was significant reduction in glucose level at 15^{th} day and 30^{th} day autopsy interval, as compared to the only irradiated mice, that is almost equal to the normal value of glucose (Control group).(Table 1)

When ANOVA test was applied it was observed that in ethanol extract treatment with days found less significant, treatment without days found moderately significant and only days found moderately significant.(Table 2)

Table 1. Variation in Glucose content of irradiated mouse with and without Opuntia elatior extract (Ethanol) treatment

Experimental Groups	Autopsy interval		
	Day1	Day 15	Day 30
Group 1 Control (without any treatment)	109.1±5.28	104.1±5.70	103.33±6.23
Group 2 Plant (Extract only)	99.33±4.26	97.5±7.44	89.0±3.82
Group 3 Plant +R (Extract-10mg/kg+6Gy Radiation)	109.5±5.27	112.33±4.71	109.83±7.75
Group 4 Radiation (6 Gy)	104±3.59	157.5±4.95	161.33±5.91

TABLE 2: Analysis of variance table for Glucose in Ethanol extract of *Opuntia elatior*

ANOVA factor	Df	Sum Sq	Mean Sq	F Value	Pr (>F)	Significance
Treatment	3	1441.42	504.81	101.29	< 2.2e48	**
Day	2	6.93	5.17	3.88	0.04451	**
Treatment : Day	6	198.77	31.18	18.643	1.15E-19	*

***Opuntia elatior* extract in Acetone**

In control group (without any treatment), the glucose level is almost constant at all autopsy intervals 1st,15th,30th days. When only plant extract (10 mg/kg body weight) were administrated to the mice, there was decrease in glucose level, which was found to be maximum at autopsy interval day 1st and reduced thereafter (107 mg/dl),that is nearby normal value of glucose (control group).When only radiation dose 6 Gy is given to the mice, in this group glucose level is increased at all autopsy intervals 1st,15th,30thday.When plant extract (10 mg/kg body weight) were administrated to mice irradiated with gamma radiations, there was significant reduction in glucose level and minimum glucose level was observed at autopsy interval 30th day(118 mg/dl),that is moderately equal to the normal glucose level(Control group). (Table 3)

When anova test applied, it was observed that in acetone extract treatment with days found less significant and treatment without days found moderately significant and only days without any treatment found highly significant. (Table 4)

Table 3. Variation in Glucose content of irradiated mouse with and without Opuntia elatior extract (Acetone) treatment

Experimental Groups	Autopsy interval		
	Day1	Day 15	Day 30
Group 1 Control (without any treatment)	108±3.8	154±5.3	109.1±7.7
Group 2 Plant (Extract only)	112.5±3.55	107.5±6.22	107±6.24
Group 3 Plant +R (Extract-10mg/kg+6Gy Radiation)	125±2.08	121.5±6.34	118.83±6.92
Group 4 Radiation (6 Gy)	133±4.93	166.66±5.28	175.5±3.31

TABLE 4: Analysis of variance table for Glucose in Acetone extract of *Opuntia elatior*

ANOVA factor	Df	Sum Sq	Mean Sq	F Value	Pr (>F)	Significance
Treatment	3	1556.45	528.33	112.36	< 2.2e56	**
Day	2	8.56	6.76	4.12	0.04892	***
Treatment : Day	6	222.21	34.19	23.338	1.15E-26	*

DISCUSSION

Njangiru et al (2017)[14] reported glucose lowering property of aqueous extract of *Psidium quajava* in alloxan induced Mice. They reported that it carried out in 3 phases; in the initial hour, the extract created a sharp reduction in blood glucose levels, followed by a steady decline from the next preceding hour. A stable elevation was then seen from the 6th hour through to the 24th hour. In the 1st hour the 4 treatment doses of the plant sample reduced blood glucose levels by 28%, 33%, 26%, and 42% respectively, in contrast to insulin which reduced blood glucose levels by 38% in same time period. In next 4th hour there was reduction in blood glucose levels by 65%, 53%, 64% and 73% in all treatment dose level respectively, compared to insulin, which reduced blood glucose levels by 74% by the similar period. At 6th hour, only the dose of 300mg/kg body weight had constant blood glucose level within normal and was comparable with insulin.

Dra *et al* (2018)[15] reported glucose lowering activity of *Caralluma europaea* against alloxan-induced diabetes in mice. According to them there was tremendous elevation in blood glucose level (262 ± 4.80 mg/dl) compared to normal group having no dose level (129.11 ± 5.22 mg/dl). The methanolic extract showed a unremitting marked decrease of blood glucose levels ($P < 0.001$) particularly 6–8–10 h after treatment. Low dose level decreased ominously the blood glucose level (from 386 ± 6.35 mg/dl to 157 ± 10.39 mg/dl at 8 h and to 87 ± 0.28 mg/dl at 10 h) in contrast with untreated diabetic mice (600 ± 0 mg/dl).

Njogu et al (2018)[16] also reported glucose reducing capacity in *Kigelia africana* (Lam). They observed that it was only at the 6th, 8th, and 24th hour there was tremendous increase in blood glucose level when compared with the initial hour ($P < .05$). There was sharp difference for all the normal group from the 2nd to the

24th hour from the initial time. Only the 0th hour, 6th, and 8th hour has difference from the 2nd hour. Significant reduction in the blood glucose levels was observed in mice having treatment dose of 50, 100, 200, and 300 mg/kg body weight of *K africana*. At dose level of 50 mg/kg, it was only at the 8th hour that significant reduces blood glucose as compared with the 0th hour ($P < .05$). Interestingly, a dose at 100 mg/kg body weight of *K africana* do not showed any difference in the blood glucose levels ($P > .05$). A dose of 200 and 300 mg/kg body weight of *K africana* only showed variations after 6, 8, and 24 hours, respectively, when compared with the normal control ($P < .05$). The 8th hour at dose of 200 mg/kg and the 24th hour for a dose of 300 mg/kg body weight of *K africana* were different from 2nd hour respectively.

Liu et al (2018)[17] proved blood sugar reducing potency of *Cyclocarya paliurus.* They reported that drug was effective after 28 days of administration to mice

CONCLUSION

Herbal drugs have been use in several traditional system of medicine for several hundred years for treating various human ailments since they often holistic treatment they are considered highly acceptable effective and non-toxic, thus treatment by *Opuntia elatior* may be useful.

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