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Comparative effect of gamma and X-ray irradiations on some characters of rice seedlings of Ashfal and Binadhan-14

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ABSTRACT

To start a mutation breeding program, it is a prerequisite to determine the effective irradiation dose rate. In this study two rice genotypes (landrace Ashfal and improved mutant variety Binadhan-14) were exposed to gamma and X-ray irradiations compare their sensitivity and to determine the effective radiation dose for mutation induction. It was found that germination percentage, seedling height and plant survival percentage decreased with the gradual increase of dose rate both in gamma and X-ray irradiation. For Ashfal, LD₅₀ and LD₃₀ were 241 Gy & 153 Gy in case of gamma ray and 215 Gy & 118 Gy in case of X-ray. In case of Binadhan-14 these values were 353 Gy & 254 Gy for gamma ray and 346 Gy & 242 Gy for X-ray. On the tested genotypes LD₅₀ and LD₃₀ were higher in gamma ray than X-ray that means a lower dose of X-ray compared to gamma ray is required to achieve same relative biological effects (RBE). These ranges of LD₅₀ and LD₃₀ values could be useful in varietal improvement program for the tested genotypes.

Key Words: Gamma ray, X-ray, LD₅₀, LD₃₀ and Rice

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I. Introduction

Rice (*Oryza sativa* L.) belongs to Graminae family and subfamily Oryzoidea is one of the most important cereal crops which have high agronomic and nutritional value that feeds more than half of the world's population. The early farmers slowly renovated *Oryza sativa* from wild rice based on their continuous selection for desirable traits. The current global production of rice is about 738.1 million metric tons per year that constitutes more than one fourth of all grain crops. Of these, Asia produced the largest part, totaling about 584 million tons (Ouma-Onyango, 2014).

Rice is the staple food crop of Bangladesh and it covers about 75% of the total cropped area, more than 80% of the total irrigated area. At present, 11.37 million hectares of land produces 34.53 million tons

of rice (BBS, 2014). Thus, rice plays a vital role in the livelihood of the people of Bangladesh. Every year cultivable land is decreasing due to the increasing population. About two million population of Bangladesh is increasing in every year and may increase by another 30 million over the next 20 years. Thus, to fulfill the requirements of increased people about 27.26 million tons more rice will be needed for the year 2020 (BRRI, 2016). To feed the bursting population it needs to increase the yield of rice through developing improved varieties. Different breeding approaches are being used for this purpose in Bangladesh. Mutation breeding is one of the effective breeding tools.

Mutation is the sudden heritable change in the genetic material in the gene or chromosome level (Chahal and Gosal, 2002). It may occur during cell division or by exposure to the DNA-damaging agents or mutagens in different environments. The effect of radiation influences by the size and weight of plants. In radiobiological reactions, the radiation dose depends on the intensity or the mode in which the total dose is fractioned. Different ionizing radiations such as gamma ray, X-ray etc. are known to influence plant growth and development through cytological, genetical, biochemical and physiological variations in cells and tissues (Gunckel and Sparrow, 1961). The radiation induces growth abnormalities in seedlings which are mainly happening due to cell damage and blocking of mitosis. In many food crops, mutation breeding has been used and becoming more popular (Mohamad et al., 2005). Of them, Mutagenesis is becoming popular in rice breeding because of its diploid nature. Ionizing radiations e.g. gamma ray, X-ray etc. are being successfully applied in inducing genetic variability in rice.

Previously, the comparative biological effectiveness and efficiency of various mutagens are useful (Smith, 1972) but not in a breeding program. Optimum dose determination is useful for varietal improvement programs (Harding et al., 2012). Many scientists attempted to determine the most effective mutagenic treatment for the induction of desirable traits in rice (Bansal et al., 1990; Katoch et al., 1992; Pillai et al., 1993; Sarawgi & Soni 1993; Kumar, 1998; Sanjeev et al., 1998; Taher et al., 2011; Harding et al., 2012). Therefore, this experiment was undertaken to assess the effectiveness and measure the effective doses of gamma and X-ray on Ashfal and Binadhan-14 to make genetic variation at seedling stage of rice.

II. Materials and Methods

Two Bangladeshi rice genotypes viz. Ashfal (landrace) and Binadhan-14 (improved mutant variety) were used in this study to observe the effect of X-ray and gamma ray irradiation. The improved mutant variety Binadhan-14 was derived from landrace Ashfal by carbon ion beam irradiation. Ashfal is photosensitive, tall, long durated and low yielding whereas Binadhan-14 is photo insensitive, semi-dwarf, short durated and high yielding rice varieties. Before irradiation, germination test was done to see the germination percentage and viability of the seeds. Twenty seeds per genotype were sown in Petri dishes containing water soaked filter paper at normal temperature and in growth chamber. Prior to mutagenic treatments, the seeds were kept in a desiccator with 60% glycerol/water mixture for seven days at room temperature for seed moisture equilibration.

Twenty pure and healthy dry seeds were irradiated with 0 Gy, 100 Gy, 200 Gy, 300 Gy, 400 Gy and 500 Gy doses of X-ray and gamma rays. For X-ray RS 2400 and gamma ray Cobalt 60 gamma irradiator GC-220 of Seibersdorf Laboratories of the International Atomic Energy Agency (IAEA), Austria was used in February, 2014. The dose rate was 12 Gy/min. and 8Gy/min. for X and gamma irradiator, respectively.

After irradiation, seeds were pre-soaked and placed between two wet papers and kept vertically in racks. Then the racks were placed in a plastic tray which contains distilled water. The bottom of blotter paper was dipped in water. Then the whole setup was placed in a plastic film-covered growth cabinet which was connected to a humidifier to maintain the humidity in the chamber (Figure 01). After seven days, data were taken on germination percentage. Then the seedlings were transplanted in the hydroponic system. Plant height was taken at seedling stage after 14 days of sowing and plant survival rate was counted at 30 days after sowing.



Figure 01. Sandwich blotter for rice germination test

Data were analyzed using Microsoft Office Excel 2007 program. Percentage plant survival was calculated using the following formula:

Percentage of survival plant = {Number of survived seedlings at 21 DAT/ Total number of seedlings transplanted at 7 DAG} × 100. LD₅₀ and LD₃₀ were calculated by regression analysis.

III. Results and Discussion

Germination percentage

Several investigations were found to work on the influence of gamma rays on seed germination of gymnosperms (Bora, 1961; Radhadevi and Nayar, 1996). The higher Exposures were usually inhibitory on seed germination of gymnosperm an Angiosperm (Thapa, 1999) whereas lower exposures were somewhat stimulator (Chauhan and Singh, 1980). Result shows that, germination percentages decreased after irradiation (Table 01). For gamma ray germination percentages started to decrease from 400Gy in Binadhan-14 but in Ashfal it decreased gradually with the increase of dose rate. In case of X-ray, germination percentages were not affected to a large extent for Binadhan-14 but it affects Ashfal. However the germination percentages decreased after irradiation, but the decrease in germination was not proportional to the increase in dosage nor was a definite pattern observed in both the varieties for gamma and X-ray irradiation. Similar results have been reported in rice by others (Harding et al., 2012; Taher et al., 2011; Cheema and Atta 2003).

Table 01. Effect of gamma and X-ray irradiation on germination of Ashfal and Binadhan-14

Dose (Gy)	Gamma ray				X-ray			
	Ashfal		Binadhan-14		Ashfal		Binadhan-14	
	Germin. (no.)	Germin. (%)	Germin. (no.)	Germin. (%)	Germin. (no.)	Germin. (%)	Germin. (no.)	Germin. (%)
0	14.67	73.33	20.00	100.0	17.00	85.00	19.00	95.00
100	14.67	73.33	19.67	98.33	13.67	68.33	20.00	100.0
200	14.33	71.67	19.67	98.33	15.67	78.33	19.00	95.00
300	13.33	66.67	20.00	100.0	15.00	75.00	19.33	96.67
400	9.33	46.67	14.00	70.00	14.67	73.33	19.33	96.67
500	9.67	48.33	12.33	61.67	13.67	68.33	18.67	93.33
Mean	12.67		17.60		14.94		19.22	
Sd	2.50		13.49		1.27		0.46	

Seedling height

Seedlings are particularly sensitive to mutagens and provide an easy means of measuring treatment effects (Kodym et al., 2011) and is widely used as an index in determining the biological effects of various physical and chemical mutagens in M₁ (Cheema and Atta, 2003; Harding et al., 2012; Konzak et al., 1972). In the tested genotypes seedling height decreased in decreasing rate with the increase of dosage and the landrace Ashfal showed more sensitivity than its progeny Binadhan-14 (Table 02). In

Ashfal 50% height reduction was observed from 300 Gy for gamma and 200 Gy for X-ray irradiation (Figure 03). For Binadhan-14 50% height reduction was observed from 400 Gy for both irradiations (Figure 02&03). Katoch et al. (1992) and Wang et al. (1995) observed a linear dependency of seedling height on the dosage of physical and chemical mutagens.



Figure 02. Seedling height of Ashfal (left) and Binadhan-14 (right) after 14 days of sowing (gamma ray treated).

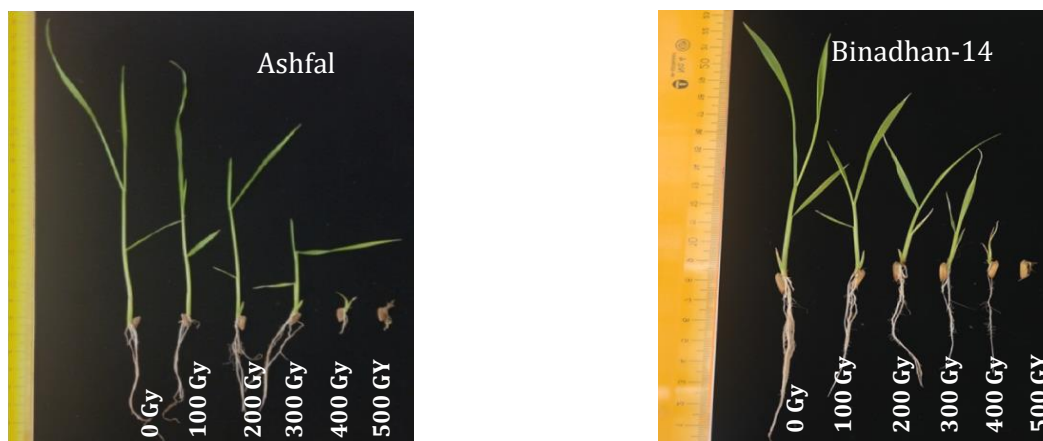


Figure 03. Seedling height of Ashfal (left) and Binadhan-14 (right) after 14 days of sowing (X-ray treated).

Table 02. The effect of gamma and X-ray irradiation on seedling height at 14 days after sowing on Ashfal and Binadhan-14

Dose (Gy)	Gamma ray				X-ray			
	Ashfal		Binadhan-14		Ashfal		Binadhan-14	
	Seedling height (cm)	% over control	Seedling height (cm)	% over control	Seedling height (cm)	% over control	Seedling height (cm)	% over control
0	17.41	100.0	12.14	100.0	18.34	100.0	13.03	100.0
100	17.36	99.72	11.65	95.90	16.07	87.61	11.34	87.06
200	13.46	77.33	10.01	82.49	9.33	50.87	11.05	84.79
300	7.64	43.88	8.10	66.73	8.22	44.83	8.70	57.30
400	0.82	4.71	3.70	52.72	3.64	19.85	7.94	48.61
500	-	-	-	-	-	-	-	-
Mean	11.34		9.66		11.12		9.84	
Sd	7.12		2.41		6.00		2.82	

Determination of lethal dose and relative biological value

The effect of gamma and X-ray irradiation on the survival percentage of Ashfal and Binadhan-14 at seedling stage in the hydroponic system after 30 days of sowing (Figure 04&05). The result shows that plant survival reduced with the increasing dose rate from 100 to 500 Gy. Physiological damage on plants became more severe above 400 Gy for Ashfal and at 500 Gy for Binadhan-14 as none of the plants survived. This result agrees with the results of [Harding et al. \(2012\)](#).

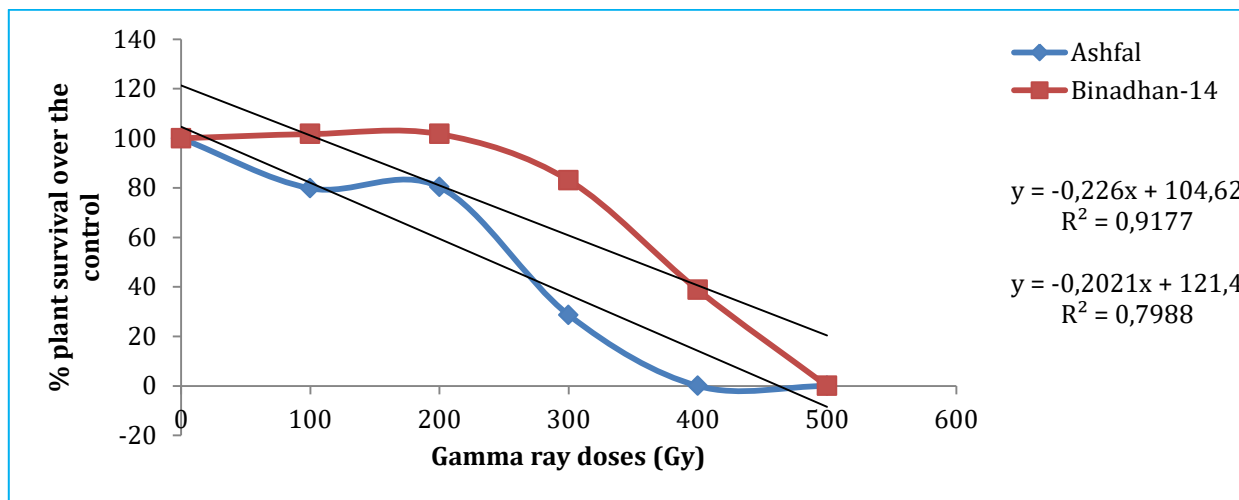


Figure 04. The effect of gamma irradiation on plant survival of Ashfal and Binadhan-14 (at 30 DAS).

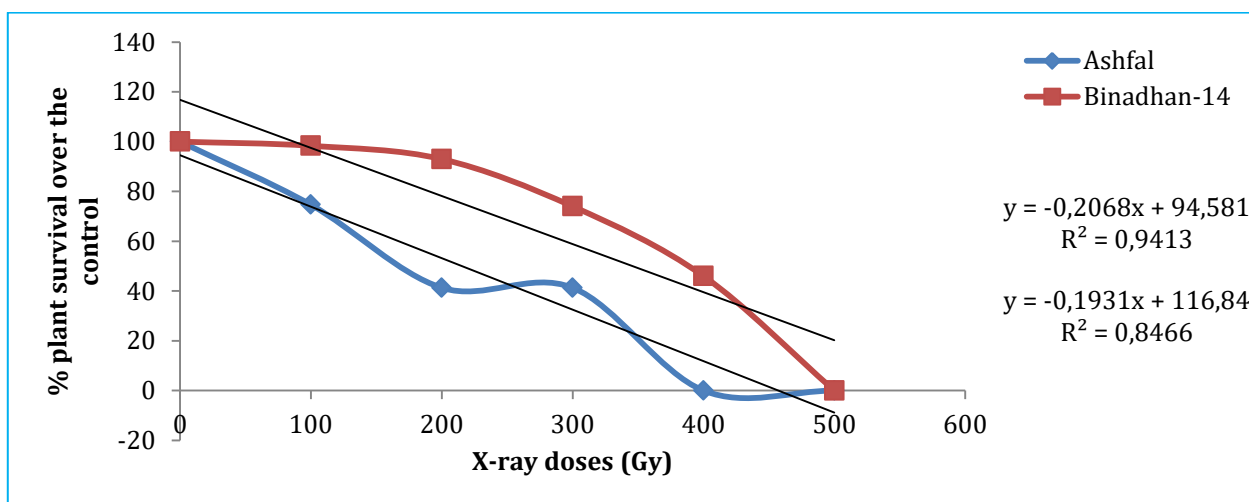


Figure 05. The effect of X-ray irradiation on plant survival of Ashfal and Binadhan-14 (at 30 DAS)

Table 03. Relative biological value (RBE) of the two irradiators for Ashfal and Binadhan-14

Variety	Gamma ray		X-ray		RBE	
	LD50	LD30	LD50	LD30	LD50	LD30
Ashfal	241.68	153.19	215.58	118.86	1.12	1.29
Binadhan-14	353.29	254.33	346.14	242.57	1.02	1.05

LD₅₀ and LD₃₀ values were determined based on percentage plant survival. For both the varieties, LD₅₀ and LD₃₀ were higher in gamma ray than the X-ray (Table 03) that means a lower dose of X-rays compared to gamma rays is required to achieve same relative biological effects (RBE). Again higher doses are required in case of Binadhan-14 to get LD₅₀ than the Ashfal that means Ashfal is more sensitive to irradiation.

IV. Conclusion

High doses of irradiation cause severe physiological damage on germination percentage, seedling height and reduce survival rate. The X-rays are more efficient than gamma rays to attain the same relative biological effects (RBE) and Ashfal was found more sensitive to irradiation than Binadhan-14. So, the calculated LD₅₀ values based on survival percentage may be useful for mutation induction and varietal improvement program of Ashfal and Binadhan-14.

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V. References

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