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Gamma Radiosensitivity Study on Chili (*Capsicum annuum*)

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Abstract: Induced mutation by gamma irradiation has been found to be a very useful technique for crop improvement. Apart from this, the proper use of induced mutation in plant breeding has become a profitable approach. This investigation was carried out to determine the LD₅₀ and effect of gamma rays on germination, plant height, survival percentage, root length, root dry weight and shoot dry weight of seedlings derived from irradiated seeds of chili (*Capsicum annuum*). Seeds of chili were treated with 300, 400, 500, 600 and 800 Gy gamma rays at Malaysian Institute of Nuclear Technology (MINT). The treated seeds including control were sown in sand beds in size 4.6 x 0.7 m² in a greenhouse at Horticulture Unit, UPMKB. Water was applied manually to maintain the soil moisture at field capacity as well as weed was manually controlled. The experiment was designed as 5 x 6 factorial in completely randomized design (CRD) with three replications. Lethal dose 50 % of population (LD₅₀) was assayed. Observation showed that germination percentage, plant height, survival percentage, root length, root weight and shoot dry weight decreased with increasing dose of gamma rays. The 800 Gy gamma ray dose had a profound effect on these variables perhaps due to injury the higher doses may have caused to the seeds of chili. This resulted in poor growth and development of chili seedlings. The LD₅₀ for chili (survival percentage) was estimated at 445 Gy. Loan contracts performance determines the profitability and stability of the financial institutions and screening the loan applications is a key process in minimizing credit risk. Before making any credit In general, higher gamma ray doses particularly 600 and 800 Gy had negative effect on the morphological characteristics of chili seedlings derived from irradiated seeds.

Key words: Gamma rays, chili, induced mutation, Malaysia

INTRODUCTION

Mutation is a change in the Deoxyribose Nucleic Acid (DNA) sequences of a gene. They may be caused by error during cell division or by exposure to the DNA-damaging agents or mutagens in the environment. One of the mutagens which have been successfully used to cause beneficial induced mutation in crops is gamma ray. Gamma rays are common used in plant breeding programmes because there are known for their simple application, good penetration, reproducibility, high mutation frequency and less disposal problems^[1]. Besides breeding, they are also used as an alternative for improvement of desired characters in agricultural crops^[2, 3, 4, 5, 6].

As a result of induced mutation and improved management and agronomic inputs over the past years, significant increase in the yield of major crops including chili varieties have reported^[4]. Chili

(*Capsicum annuum*) is an important and high demand crop in Malaysia but its cultivation has not been fully commercialized in Malaysia probably because of lack good or suitable chili varieties which adapt well to soils of this^[7,8]. But Chili has potential for commercialization in Malaysia and else where if suitable varieties are used. In this regard, induced mutation by gamma rays could be useful as with appropriate exposure doses of these ionizing radiations on chili seeds, the crop may undergo desirable mutation that could be of significant benefit to mankind. The investigation of this study was to determine the LD₅₀ and effect of gamma rays on morphological characteristics of chili seedlings derived from irradiated seeds.

MATERIALS AND METHODS

Chili (*Capsicum annuum* cv. Kulai) seeds were obtained from MARDI (Malaysia Agricultural

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Research and Development Institute). The variety was chosen because of its economic value and high germination percentage (90 %). The moisture content of the seeds was measured using Moisture Analyzer (AND MX-50) at the Agrotechnology Laboratory, Universiti Putra Malaysia Bintulu Campus (UPMBK). Seeds of chili were treated with 300, 400, 500, 600 and 800 Gy gamma rays at Malaysia Institute of Nuclear Technology (MINT). These treatments were used because doses such as 50, 100, 150, 200 and 250 Gy in our previous trial did not produce good results.

A day before planting the seeds, sand beds were prepared. The sand beds were watered to field capacity to ensure they were moist and loose enough for planting. The treated seeds were sown in the sand beds in size 4.6 x 0.7 m² in a greenhouse at Horticulture Unit, UPMKB. Water was applied manually to maintain the soil moisture at field capacity as well as weed was manually controlled. Pesticides, herbicides, and fertilizers were not applied in order to avoid any interference during the study.

Data on seed germination was recorded 13 days after planting and afterwards, 10 plants from each treatment excluding 800 Gy were transferred into polybags in a rain shelter house at UPMKB. Each polybag contained 1.2 kg blended growth medium with a top soil, sand, and processed chicken dung ratio of 3:2:1. Watering was also done daily and plant growth was monitored for 45 days. Plant height was measured weekly using a meter rule. Each measurement was carried out four times and the mean height recorded. Root length measurement was done at harvest. Roots were separated by washing (gently) away the growth medium with water. Plants were partitioned into root and shoot after which root length was determined using a ruler. Afterwards, these plant parts were bagged in brown paper bags and oven dried at 60 °C until constant weight was attained. A digital balance used to determine the dry weight of the plant parts. The plants that survived at harvest were counted. Lethal dose 50 % of population (LD₅₀) was assayed.

The experiment was designed as 5 x 6 factorial in completely randomized design (CRD) with three replications. Means in each treatment were compared by Tukey's Test using Statistical Analysis System version 9.1(SAS).

RESULTS AND DISCUSSION

The moisture content of the chili seeds was 6.08 %. This value was not consistent with the range of 12 to 14 % reported by^[11]. Table 1 shows the percentage germination of chili seed with time. There was significant interaction between dose and time. The

lower doses (300 and 400 Gy) in comparison with control (0 Gy) did not affect germination regardless of time but the contrary was the case of the higher doses (500, 600 and 800 Gy) where their effects were so pronounced that 800 Gy in particular inhibited seed germination. In the case of height, there was significant interaction between dose and time. The higher doses had significant effect on chili height such that 600 and 800 Gy in particular were lethal to the plants (Table 2). In terms of the survival of plants, irradiating chili seeds with 300 and 400 Gy had no significant effect compared with control (0 Gy) (Fig. 1). However, seeds treated with 500, 600 and 800 Gy were severely affected such that the seeds exposed to 800 Gy died while the plants derived from treating chili seeds with 600 Gy could not survive with time (Tables 1 and 2; Fig. 1). The survival for control plant was assumed as 100 % to determine the effect of the treatments applied (Fig. 1).

Compare to control (0 Gy), 600 and 800 Gy had lethal effect on root length unlike 300, 400 and 500 Gy whose effects (compared with 0 Gy) were not significant (Fig. 2). A similar observation was true for root dry weight (Fig. 3). The shoot dry weights derived from seedlings of all the seeds exposed to gamma rays were significantly lower than those not treated with gamma ray i.e. control (Fig. 4).

In terms of lethal dose (LD), the survival rate of chili was used to determine LD₅₀ and LD₁₀₀. At or above 600 Gy, the plants were unable to survive but half of the population survived (LD₅₀) at 445 Gy (Fig. 5).

The decrease in chili germination with increasing dosage could be attributed to the occurrence of seeds without completely developed embryos^[9, 10]. The main cause of germination reduction in plants has been attributed to the occurrence of seeds without completely developed embryos^[9, 10].

The fact that chili seeds which were treated with low gamma rays grew better than those exposed to higher doses, suggests that the more the seeds were exposed to gamma rays, the poorer will be the growth performance of the crop^[11] and this was so because irradiation may cause a block in cellular DNA, hence causing plant growth to stop or slow.

The survival rate of the control plants was certainly higher because their seeds were not irradiated with gamma ray. At a certain level of radiation, the plant can grow at early stage of growth but they cannot survive at certain duration probably due to DNA breakage and inability to repair them. Plants exposed to 600 Gy for example started to grow well at the early stage but they could not survive till 34 days after planting.

Table 1: Germination percentage of irradiated chili seeds with 0, 300, 400, 500, 600 and 800 Gy gamma rays after planting for 13, 20, 27, 34 and 41 days

	DAP 13	DAP 20	DAP 27	DAP 34	DAP 41	
Dosage			%			
0 Gy	42.22 ^a	44.44 ^a	43.33 ^a	43.33 ^a	42.22 ^a	43.11 ^a
300 Gy	50.00 ^a	45.56 ^a	38.89 ^a	38.89 ^a	36.67 ^a	42.22 ^a
400 Gy	43.33 ^a	38.89 ^a	31.11 ^a	31.11 ^a	31.11 ^a	35.56 ^b
500 Gy	21.11 ^b	13.33 ^b	10.00 ^b	10.00 ^b	5.56 ^b	12.00 ^c
600 Gy	15.56 ^b	2.22 ^b	1.11 ^b	0.00 ^b	0.00 ^b	3.78 ^d
800 Gy	0.00 ^c	0.00 ^b	0.00 ^b	0.00 ^b	0.00 ^b	0.00 ^d
	28.70 ^a **	24.07 ^{ab} **	21.30 ^b **	1.25 ^b **	42.22 ^a	

Means with different letter(s) in column are statistically different between treatments by the Tukey test (p = 0.05). Mean of time factor, ** Mean of dose factor. DAP means days after planting.

Table 2: Height of irradiated chili seeds with 0, 300, 400, 500, 600 and 800 Gy gamma rays after planting for 13, 20, 27, 34 and 41 days

	DAP 13	DAP 20	DAP 27	DAP 34	DAP 41	
Dosage			cm			
0 Gy	1.53 ^a	1.75 ^a	2.32 ^a	2.87 ^a	4.05 ^a	1.71 ^b
300 Gy	1.36 ^a	1.43 ^a	1.63 ^{ab}	1.77 ^b	2.37 ^b	1.63 ^b
400 Gy	1.21 ^{ab}	1.32 ^a	1.55 ^{ab}	1.75 ^b	2.33 ^b	1.14 ^c
500 Gy	0.98 ^{bc}	1.13 ^{ab}	1.12 ^{bc}	1.08 ^b	1.37 ^{bc}	0.31 ^d
600 Gy	0.74 ^c	0.50 ^{bc}	0.33 ^{cd}	0.00 ^c	0.00 ^c	0.00 ^d
800 Gy	0.00 ^d	0.00 ^c	0.00 ^d	0.00 ^c	0.00 ^c	1.71 ^b
	0.97 ^b **	1.02 ^b **	1.16 ^b **	1.25 ^b **	1.69 ^a **	

Means with different letter(s) in column are statistically different between treatments by the Tukey test (p = 0.05). Mean of time factor, ** Mean of dose factor. DAP means days after planting.

The general root and shoot dry weight decrease with increasing dosage may be attributed to poor water and nutrient utilization as poor plant growth and development leads inefficient utilization of these essential resources.

The LD₅₀ of the chili was obtained at 445 Gy while that of LD₁₀₀ was 600 Gy. The LD₁₀₀ was lower than 600 Gy but was between 500 and 600 Gy. The LD₅₀ also may vary and depends on factors such as moisture content and the variety used.

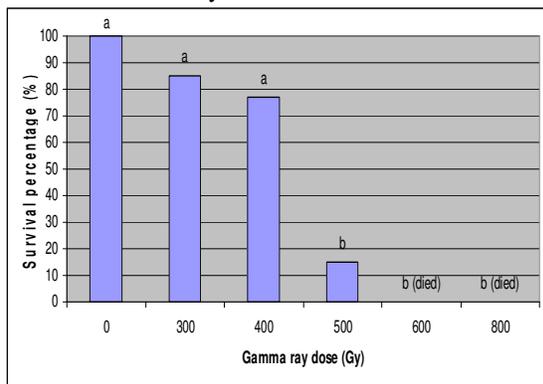


Fig. 1: Percentage survival of irradiated chili seeds with 0, 300, 400, 500, 600 and 800 Gy gamma rays after planting for 13, 20, 27, 34 and 41 days

Note: Means with different letters are statistically different between treatments by the Tukey test (p = 0.05).

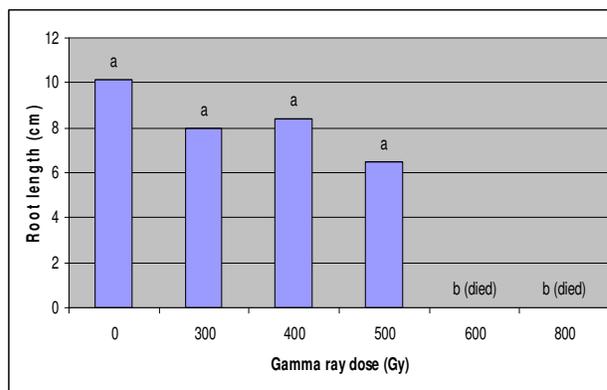


Fig. 2: Root length of irradiated chili seeds with 0, 300, 400, 500, 600 and 800 Gy gamma rays after planting for 13, 20, 27, 34 and 41 days

Note: Means with different letters are statistically different between treatments by the Tukey test (p = 0.05).

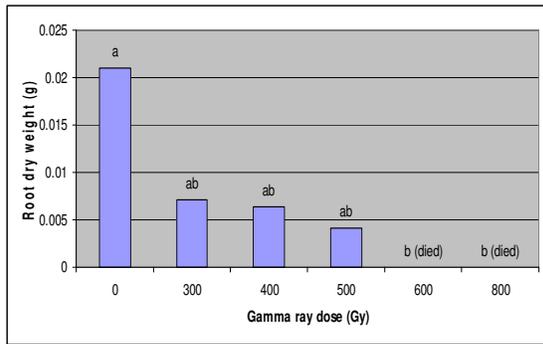


Fig. 3: Root dry weight of irradiated chili seeds with 0, 300, 400, 500, 600 and 800 Gy gamma rays after planting for 13, 20, 27, 34 and 41 days

Note: Means with different letters are statistically different between treatments by the Tukey test (p = 0.05).

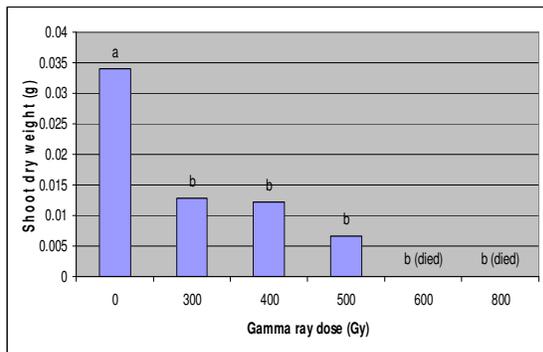


Fig. 4: Shoot dry weight of irradiated chili seeds with 0, 300, 400, 500, 600 and 800Gy gamma rays after planting for 13, 20, 27, 34 and 41 days

Note: Means with different letters are statistically different between treatments by the Tukey test (p = 0.05).

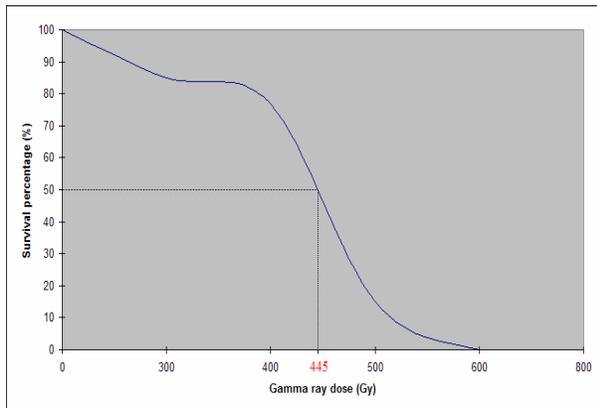


Fig. 5: LD₅₀ (50 % survival) of irradiated chili seeds with 0, 300, 400, 500, 600 and 800 Gy gamma rays after planting for 45 days

CONCLUSION

Increasing gamma ray dose decreased germination percentage, plant height, survival percentage, root length, root dry weight and shoot dry weight of chili. The LD₅₀ for survival percentage of the seedlings derived from chili seeds treated with gamma rays was obtained at 445 Gy. Generally, higher gamma ray doses particularly 600 and 800 Gy had pronounced effect on the morphological characteristics of chili seedlings derived from irradiated seeds.

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