A Study on Characteristics of Mutant Tomato (*Lycopersicum Esculentus* L.) Seed by Gamama Irradiation

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Abstract

This research deals with the effects of gamma irradiation on yield, nutritional values and antioxidant activity of tomato. The seeds from only one tomato plant of (PLATINUM 701 F1) variety were irradiated with 0.25 kGy dose of gamma ray from Co-60 and non-irradiated tomato seeds (control) were cultivated up to second generation. From the experimental results, the morphology of mutant tomato plants does not differ from that of control. The numbers of mutant tomato fruits per month (110) was found to be higher than that of control (75) during the life time. The nutritional values, some essential mineral contents (such as Na, Mg, K and Ca) of mutant tomato were nearly the same as its control. The contents of vitamin C, total phenolic, total flavonoid and antioxidant activity of mutant tomato were found to be slightly reduced from the control. The high yield of mutant tomato plant without deficiency of nutrient is achieved by gamma irradiation.

Keywords: tomato, Co-60, yield, nutritional values, antioxidant activity

1. Introduction

1.1 Nuclear Techniques in Agriculture

Gamma irradiation is often used in conjunction with other techniques, to produce new genetic lines of root and tuber crops, cereals and oil seed crops (Bagher*et al*., 2014).Induced mutation plays an important role enhancing nutritional quality in crop plants. Mutagenic agents, such as radiation and certain chemicals then can be used to induce mutations and generate genetic variation from which desired mutants may be selected (Jain, 2011). Spontaneous or induced mutation occur generally are a result of large scale deletions, inversions or translocations of chromosomes, or from point mutations (a type of mutation that causes a single change, insertion or deletion of the genetic material) in the DNA. Physical mutagens most often result in chromosome changes and larger DNA deletion while mutagenic chemicals typically cause point mutations. The degree of mutation also depends on the tissue and time and dosage of exposure. Physical mutagens, mostly ionizing radiation, can increase the natural mutation rate by 1,000 to 1 million fold, and have been widely used to induce heritable genetic changes. More than 70 percent of induced and released mutant crop varieties have been developed using physical mutagens (IAEA, 1992).

2. Materials and Methods

Firstly, the seeds from only one tomato plant of (PLATINUM 701 F1) variety were collected. 30 seeds of tomato were put into the paper bag and irradiated with 0.25kGy dose of gamma irradiation from Co-60 source which has dose rate of 0.965kGy/h at the Department of Atomic Energy, Ministry of Education. This gamma irradiated tomato seed sample and control (non-irradiated tomato seeds) were cultivated on the field scale by using transplanting method up to second generation (Figure 3.2).

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To study the variation of phenotype of mutant tomato by gamma irradiation in each generation, morphology characters (such as the plant height, leaf length, leaf width, number of leaf, stem diameter) were determined manually.

To estimate the yield and investigate the agronomical characteristics, (such as the number of flowering, numbers of fruits per plant, fruit weight, fruit length, fruiting times and life time) of mutant tomato from each generation were determined manually.

For study on nutritional qualities, the contents of moisture, ash, protein, fiber fat and carbohydrate in the mutant tomato and control from second generation were determined by air oven method, muffle furnace method, Kjeldahl distillation method, fiber cap method and soxhlet extraction method respectively. The energy value (kcal /100 g) of mutant tomato was also determined. The elemental contents (Na,Mg,K and Ca) of sample were determined by using atomic absorption spectrophotometer. The total flavonoid contents, the total phenolic contents and the antioxidant activity of ethanol and water extracts of mutant tomato and control were evaluated with spectrophotometric method using quercetin, Folin-Ciocalteu reagent and DPPH (2,2-diphenyl-1-picrylhyrazyl) radical scavenging assay. To study the variation of vitamin C content in mutant tomato during storage time, a redox titration using iodine solution (or) iodometric titration method(AOAC,2000) was used.

3. Results and Discussion

3.1 Agromorphology of Control and Mutant Tomato Plant

According to Table 2, the plant height of both GT-0 $M_1$and GT-0.25M_1were found to be 52 cm in first generation and in second generation, the plant height was found to be 47 cm for GT-0 M_2 and 49 cm for GT-0.25M_2. Therefore, the height of mutant tomato plants in each generation did not differ from their control.

According to the experimental results, the shape of leaves of mutant tomato plant did not change due to gamma irradiation in each generation (Figure 3.1). The leaf length and width of mutant plants GT-0.25were found to be 16 cm and 11 cm in first generation and 15 cm and 11 cm in second generation whereas leaf length and width of GT-0were found to be 19 cm and 12 cm and18 cm and 13 cm in each generation respectively. The number of branches and the stems diameter of mutant tomato were found to be the same as the control. The number of leaves of mutant tomato plant was found to be slightly higher than that of control. According to the overall results, mutant tomato plants did not significantly change from their control.

Table 1 Agromorphology of Control and Mutant Tomato Plant

Agronomical Parameters	Samples			
	First generation		Second generation	
	GT-0 M_1	GT-0.25 M_1	GT-0 M_2	GT-0.25 M_2
Plant height (cm)	52.0± 9.2	52.0 ± 2.8	47.0 ± 3.3	49.0 ± 1.7
Leaf length (cm)	19.0± 0.9	16.0 ± 1.7	18.0 ± 0.3	15.0 ± 1.7
Leaf width (cm)	12.0± 0.5	11.0 ± 1.2	13.0 ± 1.4	11.0 ± 1.0
Stem diameter (cm)	0.9± 0	0.8 ± 0.2	0.8 ± 0.1	0.8 ± 0
Numbers of leaf	12.0± 1.9	15.0 ± 1.5	10.0 ± 1.0	13.0 ± 1.1
Numbers of branch	2.0 ± 0.5	2.0 ± 0	2± 0.5	2.0 ± 0

GT-0 M_1 = mutant tomato gamma irradiated with 0 kGy (control) in first generation

GT-0.25M_1 = mutant tomato gamma irradiated with 0.25 kGy in first generation

GT-0 M_2 = mutant tomato gamma irradiated with 0 kGy in second generation

GT-0.25M_2 = mutant tomato gamma irradiated with 0.25 kGy in second generation

3.2 Yield and Agronormical Characteristics of Control and Mutant Tomato Plant

To study the estimation on yield, the experimental results (Table 3) revealed that the number of fruits per plant of GT- 0 $M_1$and GT-0.25 M_1 were 12 and 31 respectively. The number of fruits per month was found to be 121 for GT- 0 M_1 and 148 for GT-0.25 M_1 during the life time of tomato in first generation. In second generation, the number of fruits per plant of GT-0 M_2 and GT-0.25 M_2 were 14 and 19 respectively. The total number of fruits of control was found to be 75 and 110 for GT-0.25 M_2 within one month. The fruit weight of mutant tomato was slightly increased from its control in each generation(Figure 3.3). To sum up, mutant tomato plant in GT-0.25 M_2 case provide higher yield than that of control.

Table 2 Yield and Agronormical Characteristics of Control and Mutant Tomato Plants in First Generation

Agronomical Parameters	Samples			
	First generation		Second generation	
	GT-0 M_1	GT-0.25 M_1	GT-0 M_2	GT-0.25 M_2
Number of fruit per plant	12.0± 0.5	31.0 ± 0.4	14.0 ± 0.5	19.0 ± 0.6
Number of fruit per month	121.0±2.9	148.0± 2.3	75.0±2.0	110.0 ± 2.2
Fruit weight (g)	20.0± 1.3	31.0 ± 2.2	35.0± 2.3	36.0 ± 2.1
Fruiting times	4.0	3.0	4.0	4.0
Life time (day)	96.0	110.0	98.0	99.0

GT-0 M_1 **GT-0.25 M_1** **GT-0 M_2** **GT-0.25 M_2**

Fig 3.1Harvested Tomatofruits of control and mutant tomato plant in first and second generations

3.3 Nutritional Values of Control and Mutant Tomato in Second Generation

The nutritional values of GT-0 M_2 and GT-0.25 M_2 were determined by AOAC methods. From Table (4), the moisture content of GT-0 $M_2$and GT-0.25 M_2 were found to be 95.24 %, and 94.97 % . The ash contents were 0.61 % and 0.57 %. The protein contents were 0.07 % and 0.06 %. The fiber contents were 1.32 %, and 1.61 %. The fat contents were 0.06 % and 0.08 % and the carbohydrate contents were 2.60 % and 2.67 % respectively. The energy values of these samples were 11.22 and 11.40 kcal/100g.Therefore, from the overall results, the nutritional values of mutant tomato are nearly the same as the control.

Table 3 Nutritional Values of Control and Mutant Tomato in Second Generation

No.	Nutritional Parameters (%)	Samples	
		GT–0 M_2	GT–0.25 M_2
1	Moisture	95.24	94.97
2	Ash	0.61	0.57
3	Protein	0.07	0.06
4	Fiber	1.32	1.61
5	Fat	0.06	0.08
6	Carbohydrate	2.60	2.67
7	Energy value (kcal/100g)	11.22	11.40

3.4 Elemental Contents of Control and Mutant Tomato in Second Generation

According to the experimental results (Table 5),the content of sodium in control and mutant GT-0.25 M_2 was found to be 0.40 and 0.39 mg /100 g. The contents of magnesium in GT-0 M_2 (control) and mutant GT-0.25 M_2 were found to be 0.13 and 0.14 mg/100g respectively. From this result, some nutrient elemental contents (Na, Mg, K and Ca) of mutant tomato were found to be significantly unchanged from that of control.

Table 4 Elemental Contents of Control and Mutant Tomato in Second Generation

No.	Samples	Elemental Contents (mg/100g)			
		Na	Mg	K	Ca
1	GT-0 M_2	0.40	0.13	0.21	0.55
2	GT-0.25 M_2	0.39	0.14	0.21	0.56

3.5 Vitamin C Contents of Control and Mutant Tomato in Second Generation

From Table 6, it was found that the contents of vitamin C in GT-0 M_2 (control) and GT-0.25 M_2 were found to be 20.82 and 19.18 mg/100 g respectively. It was found that the vitamin C content of GT-0.25 M_2 was slightly reduced than that of control.

Table 5 Vitamin C Contents of Control and Mutant Tomatoin Second Generation

No.	Samples	Vitamin C content *(mg/100 g)
1	GT-0 M_2	20.82
2	GT-0.25 M_2	19.18

3.6 Total Phenolic and Total Flavonoid Contents of Control and Mutant Tomato in Second Generation

The phenolic compounds are a large class of plant secondary metabolites. They are important for the quality of plant based foods and they are responsible for the colour of red fruits, juices and wines. The phenolic compounds, particularly flavonol, are known to be potent free radical scavengers and antioxidant (Sahelian, 2014). From Table 7, the total phenolic content of ethanol extract of GT-0 M_2 (control) and mutant GT-0.25 M_2 were found to be 76.86 and 65.90 g GAE/mg of extract and water extract of these samples were found to be 73.32 and 72.02g GAE/mg of extract. The total phenolic contents of mutant GT-0.25 M_2 samples was slightly reduced than that of control in both ethanol and water extracts.

Flavonoid is a group of natural substances with variable phenolic structures. This is attributed to their anti-oxidative, anti-inflammatory, anti-mutagenic and anti-carcinogenic properties coupled with their capacity to modulate key cellular enzyme function (Atmani *et al*.,2009). The total flavonoid content of ethanol extract of GT-0 M_2 (control) and mutant GT-0.25 M_2 were found to be 4.75 and 2.25 g QE/ mg of extract. The total flavonoid contents of water extract of these samples were found to be 8.50 and 6.00 ag QE/ mg of extract. It was found that the total flavonoid content of both ethanol and water extracts of GT-0.25 M_2 was slightly reduced than that of control.

Table 6Total Phenolic and Total Flavonoid Contents of Control and Mutant Tomato in Second Generation

Samples (100 g/mL)	Total Phenolic Content (µg GAE/ mg of extract)		Total Flavonoid Content (g QE/ mg of extract)	
	Ethanol Extracts	Water Extracts	Ethanol Extracts	Water Extracts
GT-0	76.86	73.32	4.75	8.50
GT-0.25 M_2	65.90	72.02	2.25	6.00

3.7 Antioxidant Activities of Control and Mutant Tomato in Second Generation

From the experimental results (Table 8) revealed that IC_{50} values of ethanol extracts of GT-0 M_2 and GT-0.25 M_2 samples were 17.49 and 22.50 g/mL and IC_{50} values of water extracts of GT-0 M_2 and GT-0.25 M_2 samples were 11.79 and 19.23 g/mL. Based on the results, 0.25 dose of gamma irradiation is found to be slightly reduced in antioxidant activity of tomato due to slightly reduce the vitamin C and total phenolic content of it. Nevertheless, it cannot affect the risk of health for human because the reducing amount is not significantly large scale.

Table 7 Antioxidant Activities of Control and Mutant Tomato in Second Generation

No.	Samples	IC_{50} (µg/mL)	
		Ethanol Extracts	Water Extracts
1	GT-0 M_2	17.49	11.79
3	GT-0.25 M_2	22.5	19.23
4	Ascorbic acid	0.74	

CONCLUSION

In this research, effects of gamma irradiation on yield, morphology, the nutritional values, some essential mineral contents, the vitamin C, the total phenolic content, the total flavonoid content and the antioxidant activity of mutant tomato are found out that: the morphology is not different from that of control. The mutant tomato plants in GT-0.25 M_2 provide more numbers of fruits than that of control. It provides higher yield with same nutritional values, essential mineral contents and slightly reduces the vitamin C, total phenolic, the total flavonoid and the antioxidant activity than that of control. This reducing amount does not affect the risk of human health because it is not a significantly large scale. Therefore in this research, the increase yield of mutant tomato plant without deficiency of nutrient is achieved by gamma irradiation.

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References

AOAC. (2000). "Official Methods of Analysis". Association of Official Analytical Chemists, Manual of Food Quality Control, Washington:17th Ed., 714

Atmani, D., N. Chaher, H. Boudaoud, H. Debache and M. Berboucha. (2009). "Flavonoids in Human Health: from Structure to Biological Activity". *Curr. Nutr. Food Sci.,* **5**, 225-237

Bagher, A. M., N. Askari, M. Mirhabibi and V. Mirzaei. (2014). "Nuclear Techniques in Agriculture and Gentics". *American Journal of Bioscience,* **2**(3), 102-105

IAEA. (1992). "Plant Breeding: Induced Mutation Technology for Crop Improvement". *Units at the IAEA's Seibersdorf Laboratories*, **4**, 25-27

Jain, S. M. and P. Suprasanna. (2011). "*Induced Mutation for Enchancing Nutrition and Food Production*". Nuclear Agriculture and Biotechnology Division, **40**, 201-215

Sahelian, R.M.D. (2014). " Phenolic Compounds from Plant Foods " . *Phytochemistry Review*,**11**(2),153-177