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Effects of Gamma Radiation on Germination, Growth Characteristics and Morphological Variations of *Moluccella laevis* L.

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Abstract: In the present study, seeds of *Moluccella laevis* L. were divided into two parts, one part were soaked for 12 hours in water before treatments, while the other were used as a dried. Seeds were exposed to gamma rays at doses (0, 2.5, 5, 7.5, 10, 12.5,15, 17.5 and 20 Kr) to examine their effects on germination and survival percentage, growth traits and morphological variation. The highest germination percentage was obtained by 2.5 and 5 Kr of dry seeds and 2.5 Kr of wet seeds in both seasons. Low doses of wet seeds and all doses of dry seeds except 20 Kr had the same plant survival percentage 100% in both seasons. Low dose of wet seeds increased plant height, while higher doses decreased it. Higher gamma radiation doses of wet seeds increased number of branches and dry weight of vegetative growth. The doses of 7.5, 17.5 and 20 Kr of dry seeds and 5 Kr of wet seeds were hastened flowering date. Low dose (2.5 Kr) of dry and wet seeds increased number of flowers per branches. Wet treatments caused a simulative effect in most characters. The high doses 12.5 to 17.5Kr of wet seeds caused some morphological variations. The genetic relationship of the morphological variations can be determined by using RAPD analysis.

Key words: *Moluccella laevis* L. · Gamma rays · Germination percentage · Morphological variations · RAPD analysis

INTRODUCTION

Gamma rays (is a part of electromagnetic spectrum) belong to ionizing radiation can be energetically charged particles, such as electrons, or high-energy photons. The biological effect of gamma rays is based on the interaction with atoms or molecules in the cell, particularly with water to produce free radicals in cells. These radicals can damage or modify important components of plant cells and have been reported to affect differentially the morphology, anatomy, biochemistry and physiology of plants depending on the irradiation level. These effects include changes in the plant cellular structure and metabolism, e.g., dilation of thylakoid membranes, alteration in photosynthesis, modulation of the antioxidative system and accumulation of phenolic compounds [1, 2]. The primary effects of ionizing radiation are ionization, dissociation and excitation. The excitation cause a weak interaction and the ionization and dissociation resulted in strong interaction. Absorption of ionizing radiation in biological materials, there is a possibility that it will act directly on critical targets in the cell [3]. The effects observed after exposure were deeply influenced by several factors, some related to plant characteristics (e.g., species, cultivar, stage of development, tissue architecture and genome organization) and some related to radiation features (e.g., quality, dose, duration of exposure) [4].

A number of radiobiological parameters are commonly used in early assessment of effectiveness of radiation to induce mutations. Mutagenic radiations are a useful tool to all breeding programs, thus in the flowering crops [5]. Gamma radiation can be useful for the alteration of physiological characters [6]. There are several usages of nuclear techniques in agriculture. In plant improvement, the irradiation of seeds may cause genetic, variability that enable plant breeders to select new genotypes with improved characteristics such as precocity, salinity

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tolerance, grain yield and quality [7]. The irradiation of seeds with high doses of gamma rays disturbs the synthesis of protein, hormone balance, leaf gas-exchange, water exchange and enzyme activity [8]. Due to limited genetic variability among the existing plant genotypes, opened a new era for crop improvement and now mutation induction has become an established tool in plant breeding that can supplement the existing germplasm and can improve cultivars in certain specific traits as well [9].

Moluccella laevis L) (x= 34) [10] Family Lamiaceae order Lamiales is annual ornamental plant with leafy spikes of cuplike green calyxes surrounding small, white, fragrant corollas within. The flowers are two-lipped and tubular. Its common names are shell flower and Bells of Ireland. It can be used as annual borders and cut flower. The actual flower is a small white fragrant flower inside the "bell." When dried, the leaves turn pale beige and will last for years [11].

PCR-based DNA marker techniques seem to provide the means for generating useful information on polymorphism, genetic relatedness and diversity. The PCR-based random amplified polymorphic DNA (RAPD) markers are dominant markers and are extensively used in genetic mapping [12] and for the identification of markers linked with useful traits [13]. Due to its technical simplicity and speed, RAPD methodology has been used for diversity analyses in several crops [14].

The main objective of the present investigation was to study the effects of different doses of gamma rays from Cobalt -60 on seeds germination and the possibility of inducing additional variation in *Moluccella laevis* L. which can be more cut flower value.

MATERIALS AND METHODS

Plant Materials: Seeds of local variety of *Molucella laevis* L. were used in this experiment. Seeds were divided into two parts, one part was soaked for 12 hours in water before treatments, while the other was used as a dried treatments.

Gamma Irradiation: Gamma – rays used in this study were generated from the Cobalt-60 source, in Gamma Cell 3500, installed in the Irradiation Laboratory at Middle East Regional Radio – Isotope Center for the Arab Countries at Dokki, Giza, Egypt. The used doses were 0; 2.5; 5.0; 7.5; 10.0; 12.5; 15.0; 17.5 and 20.0 kr. The dose rate was 1.02 rad/ sec. in the first season (2009/2010) and 0.86 rad/sec in the second one (2010/2011).

Experiment: The experiment was carried out during two seasons (2009/2010 and 2010 /2011) in the Floriculture and Ornamental Horticulture Research Branch, Antoniadis Garden, Alexandria, Egypt. Treated seeds of gamma rays were sown on 4 October 2009 and 12 October 2010 in the first and second seasons, respectively in 9 cm pots containing a mixture of 1:1 by volume of sand and clay. After five weeks from sowing, the seedlings were transplanted in 25 cm diameter clay pots. The experiments were terminated on 5 May 2010 and 15 may 2011 in the first and second season, respectively.

Data: Germination percentage of M_1 was calculated as follows: 100 seeds from each treatment were sown in Petri dishes containing each 5 ml of distilled water. Petri dishes were placed in an incubator for 6 days at 25°C. Number of germinated seeds was recorded after 10 days. The final germination percent (FGP) was calculated the formula:

(FGP) = (Number of germinated seeds after 10 days / Total number of seeds) X 100

With the onset of the first flower, flowering date was recorded. With open fifty percent of the flowers on plant; plant height, number of branches/ plant, vegetative growth dry weight and number of flowers/plant were recorded.

Statistical Analysis: The statistical layout of the experiment was split-plot design with 3 replicates and 9 treatments with 15 plants per plot. The main plot represented the seeds dry and wet treatments, while the sub plot described the gamma doses. The mean and ANOVA were calculated using SPSS (version 10) software. The mean separations were carried out using Duncan's multiple range tests [15] and significance was determined at $p < 0.05$.

Plant DNA Extraction and RAPD-PCR Condition: All plants of the different treatments were examined daily to isolated the variations on the basis of vegetative growth and panicles which concluded leaf changes (color and shape), branching and flowering changes of each treatment to determine the accessions which exhibited abnormal phenotypes as following Table (1).

Young leaves were taken from each plant and thoroughly washed with water then ethanol to remove dust and other contaminants. The DNA was extracted using Fermentas Plant tissue DNA purification kit.

Table 1: The Abnormal phenotypes of M1 generation in both seasons

Abnormal phenotype	Dose	No.
Control	Control	1
Variegated plant	15 Kr of Wet seeds in the second season	2
Leaf shape	12.5Kr of Dry seeds in the first season	3
Flower shape	12.5Kr of Dry seeds in the first season	4
Tall plant	17.5 Kr of Dry seeds in the first season	5
Dwarf plant	20 Kr of Wet seeds in the first season	6

Table 2: RAPD primers used in the study.

No of bands	Sequence5'-3'	Primer
8	GGTGCGGGAA	RAPDA1
6	GTTTCGCTCC	RAPDA2
8	GTAGACCCGT	RAPDA3
8	AAGAGCCCGT	RAPDA4
8	AACGCGCAAC	RAPDA5
9	CCCGTCAGCA	RAPDA6
10	AACTTACCGC	UBC 162
10	CGGTGACATC	UBC 232
8	TGAGCGGACA	OPD 05

The DNA was quantified spectrophotometrically and by electrophoresis on 0.8% agarose gels. PCR reactions were conducted using 9 primers (10-mer primers) (GENT Bio) with the following sequences Table 2.

Total Genomic DNA of each accession was diluted in sterile double distilled water to a concentration of 10 ng/µl for RAPD analysis. PCR was performed in PEQLAB thermocycler, Germany in a 25 µl reaction volume containing the following reagents: 1.0 µl of dNTPs(10mM), 1.0 µl of $MgCl_2$ (25mM), 5µl of 10x buffer, 1.0 µl of primer (10 pmol), 1.0 µl of DNA (25 ng/ µl), 0.3 µl of taq polymerase (5µ/µl) and 15.7 ddH_2o.. After initial heating for 5 min at 94°C, samples were PCR amplified using 40 cycles (94°C, 20 sec; 42°C, 20 sec; 72°C, 1 min) followed by a final extension of the PCR products for 4 min at 72°C. The products of amplification were analyzed by electrophoresis in 2.0 % agarose gels with TAE running buffer, visualized by ethidium bromide staining and photographed under UV light with a digital Sony Cybershot camera. Each reaction was repeated twice and negative controls accompanied the reactions without adding DNA for increasing the fidelity of the data.

Data Analysis: All visible and unambiguously scorable fragments amplified by the primers were scored by visual observation. Amplification profiles (band in each position) were scored as present (1) or absent (0). The scores obtained using primers in the RAPD analysis were then joined and used to estimate genetic distances using Jaccard coefficient [16] in a computer program, RAP Distance [17]. UPGMA (Unweighted Pair Group Method of Arithmetic Means) dendrogram was created using genetic distances in PAUP4 [18].

RESULTS AND DISCUSSION

Effect of Gamma Irradiation on Seed Germination and Plant Survival: 1.1. Seed germination: The results revealed significant effects of radiation doses on germination percentage (Table 3). The low doses of gamma radiation increased the germination percentage when compared with control and the higher doses. The highest germination percentage was o btained by 2.5 and 5 Kr of dry seeds and 2.5 Kr of wet seeds in both seasons. Also, soaking seeds in water increased the negative effect of higher gamma rays doses on germination percentage, whereas the dose 20 Kr of wet seeds gave the lowest germination percentage in the first season and prevented the germination in the second one. Acceleration of seed germination by low doses of ionizing radiation has been attributed to many factors. Maherchandani [19] attributed the promotion of *Avena fatua* L. seed germination to the increase in oxygen uptake following irradiation with low doses of gamma rays, which resulted in the production of organic and inorganic peroxy radicals, which led to breaking seed dormancy. Also, the stimulating causes of gamma ray on germination may be certified to the activation of RNA or protein synthesis, which occurred during the early stage of germination after seeds irradiated [20]. On the other hand, the inhibition of seed germination at high doses could be due to the damage in seed tissue, chromosomes and subsequent mitotic retardation and the severity of the damage depend on the doses used [21, 22].

Plant Survival: The sensitivity of dry and wet *Molucella laevis* seeds to gamma radiation was evaluated by comparing the survival rate (%) among low doses, higher doses and non-irradiated plantlets. Highly significant differences were observed among the low and higher doses of irradiation (Table 3). The results showed that control, low doses (2.5, 5, 7.5 and 10 Kr) of wet seeds and all doses of dry seeds except 20 Kr had the same plant survival percentage 100% in both seasons. On other hand, the higher doses (12.5, 15, 17.5 and 20 Kr) of wet seeds liner decreased the plant survival percentage with the increase of gamma radiation doses in both seasons.

Table 3: Effect of gamma radiation of dray and wet seeds on seeds germination and plant survival percentage of *Molucella laevis* L. in both seasons

Plant survival %		Seeds germination %			
Second season	First season	Second season	First season	Dose (Kr)	Treatment
100 a	100 a	84.7 cd	82.3 c	0	Dry seeds
100 a	100 a	93 a	90 ab	2.5	
100 a	100 a	94 a	92 a	5	
100 a	100 a	90 b	88 b	7.5	
100 a	100 a	85.7	71.6 e	10	
100 a	100 a	80 d	76.6 d	12.5	
100 a	100 a	60 g	50 h	15	
100 a	100 a	75 e	81.6 c	17.5	
73.33 c	93.33 b	60 g	41.6 g	20	
100 a	100 a	85.7 c	88.3 b	0	Wet seeds
100 a	100 a	93.4 a	91 ab	2.5	
100 a	100 a	88 bc	79 cd	5	
100 a	100 a	86 c	82 c	7.5	
100 a	100 a	75 e	60 f	10	
86.6 b	80 c	72 f	58 f	12.5	
53.3 d	60 d	42 h	39 g	15	
46.6 e	40 e	44 h	41 g	17.5	
0 f	33.33 f	0 i	10 i	20	
**	**	**	**	Sig.	

Means of treatments in the column have the same letters, are not significantly different at 5% level according to Duncan's multiple range test.

These results were in accordance with the germination test done by Moghaddam *et al.* [23] on *Centella asiatica*, they noted that the plantlet survival rate kept decreasing with increasing irradiation dosage for three weeks after irradiation. The results obtained by Kiong *et al.* [6] indicated that survival of plants to maturity depends on the nature and extent of chromosomal damage. Increasing frequency of chromosomal damage with increasing radiation dose may be responsible for less germinability and reduction in plant growth and survival. These results are in agreement with those obtained by Park *et al.* [24] on *Hosta plantaginea* and Golubinova and Gecheff [25] on Sudan grass.

Effect of Gamma Radiation on Vegetative Growth

Plant Height: From the obtained data noticed that gamma ray had a highly significant impact on plant height (Table 4). The tallest plants were observed on the low doses of gamma rays (2.5, 5 and 7.5 Kr) of wet seeds in both seasons when compared with control and low doses of dry seeds. On contrast, by increasing radiation dose to 12.5, 15, 17.5 and 20 Kr of wet seeds in both seasons decreased the plant height. The maximum deceased in plant height (46.33 and 50. 83 cm) was observed when wet seeds were exposed to 20 and 12.5 Kr in the first and second season, respectively. There are many explanations for the stimulatory effects of low-doses of gamma radiation, Wi *et al.* [26] noted that there is a hypothesis that the low dose irradiation will induce the growth stimulation by changing the hormonal signaling network in plant cells or by increasing the anti oxidative capacity of the cells to easily overcome daily stress factors such as fluctuations of light intensity and temperature in the growth condition. In contrast, the high-dose irradiation that caused growth inhibition has been ascribed to the cell cycle arrest at G2/M phase during somatic cell division and/or various damages in the entire genome [27]. Similar results were obtained by El-Ashry *et al.* [28] on *Lathyrus odoratus* and El-Mahrouk [29] on *Gomphrena globosa.*

Number of Branches and Dry Weight of the Vegetative Growth: The results showed that gamma radiation gave a highly significant effect on number of branches and dry weight of the vegetative growth (Table 4). High doses of gamma rays (17.5 and 20 Kr) of wet seeds increased number of branches in both seasons when compared with control. While, the radiation of dry seeds not affected significantly on number of branches. Also, seedlings irradiated with 15 and 17.5 Kr of wet seeds gave the

Table 4: Mean values of the plant height (cm), number of branches & Dry weight of vegetative growth (g) of *Molucella laevis* L. derived from irradiated of dried and wet seeds by different gamma ray doses during the two seasons

Dry weight of vegetative growth (g)		Number of branches		Plant height (cm)			
Second season	First season	Second season	First season	Second season	First season	Dose (Kr)	Treatment
17.81cd	15.95c	5.45ab	4.86b	68.69c	65.58c	0	Dry seeds
13.49de	11.78de	4.51b	3.93bc	68.78c	65.66c	2.5	
24.06bc	21.23bc	4.88ab	4.30b	66.78cd	63.66cd	5	
24.15b	21.00bc	4.43b	3.85bc	69.53bc	66.41c	7.5	
15.58d	13.64cd	4.55ab	3.96bc	56.84f	54.77e	10	
11.66e	10.25de	4.66ab	4.08bc	57.89f	53.72e	12.5	
8.87f	8.10e	4.81ab	4.23b	62.33e	59.22d	15	
21.39bc	18.45bc	4.16b	3.58bc	57.64f	54.52e	17.5	
17.90cd	16.02c	5.43ab	4.89b	66.09d	62.97d	20	
24.37b	22.31b	4.95ab	4.53b	71.11b	69.11bc	0	Wet seeds
23.70bc	21.8b	5.20ab	4.78b	73.05 ab	70.94a	2.5	
19.51c	17.66c	4.43b	4.01bc	73.72 a	71.72a	5	
20.55c	18.74bc	4.76ab	4.35b	72.94 ab	71.05a	7.5	
9.49ef	9.15e	4.68ab	4.26b	67.26 cd	65.26cd	10	
10.05ef	9.34e	3.41b	3.00c	50.83 g	48.83f	12.5	
30.60a	28.24a	5.41ab	4.99ab	63.77 e	60.44d	15	
30.57a	28.32a	5.66a	5.25a	62.44e	61.78d	17.5	
-	20.25bc	-	5.19a	-	46.33f	20	
**	**	**	**	**	**	Sig.	

Means of treatments in the column have the same letters, are not significantly different at 5% level according to Duncan's multiple range test.

Table 5: Effect of irradiated dry and wet seeds by gamma rays of *Molucella laevis* L. on number of days to flowering and number of flowers per branch during two seasons

Number of flowers per branch		Number of days to flowering			
Second season	First season	Second season	First season	Dose (Kr)	Treatment
97.68 cd	53.63 c	118.06 d	116.90 d	0	Dry seeds
63.15 ef	62.73 ab	126.46 bc	122.63 c	2.5	
65.80 ef	54.54 bc	123.80 cd	125.30 bc	5	
54.94 fg	58.80 b	120.23 d	119.06 d	7.5	
52.74 g	49.94 cd	131.16 ab	131.60 a	10	
68.08 e	48.41 d	132.76 a	130.00 ab	12.5	
64.50 ef	57.01 bc	126.76 bc	125.33 bc	15	
61.88 ef	57.17 bc	120.50 d	120.60 d	17.5	
97.68 cd	58.21 b	119.60 d	118.43 d	20	
91.02 d	55.37 bc	126.03 bc	125.20 bc	0	Wet seeds
123.3 a	63.55 a	129.23 b	127.43 b	2.5	
102.36 c	59.83 ab	120.26 d	119.40 d	5	
101.43 c	61.76 ab	128.50 bc	125.66 bc	7.5	
106.03 bc	60.13 ab	126.50 bc	127.66 b	10	
105.8 bc	54.63 bc	125.77 c	119.70 cd	12.5	
109.96 b	54.85 bc	131.56 a	130.73 ab	15	
108.23 bc	52.24 cd	125.53 bc	124.93 bc	17.5	
-	52.19 cd	-	126 .00 b	20	
**	**	**	**	Sig.	

Means of treatments in the column have the same letters, are not significantly different at 5% level according to Duncan's multiple range test

highest dry weight of vegetative growth in both seasons. Similar results were reported by Khangyldin [30] who suggested that an increase in kinetin to auxin increased buds, leaves and shoots. The same hormonal balance may also be formed by gamma irradiation or kinetin. The production of growth hormone, kinetin have been stimulated which resulted in increased number of branches and leaves. While, the increase in vegetative growth weight could be attributed to increase plant height, branches and leaves number. These results are in agreement with those obtained by El-Ashry et al.[[28] on *Lathyrus odoratus* Koli *et al.* [31] on Cumin.

Effect of Gamma Radiation on Flower Date and Number of Flowers per Branch: Data presented in Table 5 indicated that the doses of gamma rays caused significant effects on flower date and number of flowers. The doses of 10 and 12.5 Kr of dry seeds and 15 Kr of wet seeds produced

progressive delay flowering in the first and second season, respectively. On contrast, the doses of 7.5, 17.5 and 20 Kr of dry seeds and 5 Kr of wet seeds were hastened flowering date in both seasons and had the same trend like control of dry seeds. As regards to the interaction between the dose and soaking there are significant effects in both seasons, higher doses of dry treatments and lower doses of wet treatments had the same results to decreased the number of day to flowering. In respect of number of flowers per branch data show that low dose of gamma rays (2.5 Kr) of dry and wet seeds increased number of flower per branch in both seasons, on contrast with increasing the doses caused negatively effect on flower number per branches. The higher efficiency of a lower concentration of mutagenic agent is due to the fact that biological damage (seedling injury, lethality and sterility) increased with increasing in dose at faster rate than the mutations [32]. Inhibition effect by gamma-irradiation may be related to auxin and DNA biogenesis in a relationship which shown that (1) DNA is required for and is previously synthesized sequentially to auxin formation, the radiation block occurring in the formation of nucleic acid; (2) that the primary radiation block is in auxin synthesis, the auxin required for the formation of DNA; and (3) that the effect of radiation is on an undefined entity in reaction previous to and essential for both DNA and auxin synthesis [4,33, 34]). These results are in agreement with those obtained by Abdel- Maksoud [35] on *Solanum pseudocapsicum* and Tokar and Cagirgan [36] on Kabuli chickpea.

Effect of Radiation on the Induction of Variations: Figure 1 showed that there were abnormal phenotypes observed by gamma rays treatments. Irradiated wet seeds resulted of an increase in number of branches and some leaf abnormalities by 17.5 Kr, variegated leaves by 12.5 & 15 Kr and calyx abnormalities by 20 Kr. While, irradiated dry seeds by 7.5 Kr resulted of abnormal spick. These results are in agreement with those obtained by Venkatachalam and Jayabalan [37] they found on *Zinnia elegans* cv. crimsom red that 7.5 Kr gamma rays produced significant more morphological changes. Dhankhar and Dhankhar [38] determined the effects of different doses of gamma- rays at 0.6 and 0.7 kr. on okra red line MR-54-2 seeds. They indicated that the size of leaves decreased with increasing gamma radiation dose.

Random Amplified Polymorphic DNA (RAPD) Analysis: The obtained results for the RAPD analysis showed that all primers were proven to be polymorphic, produced 75 bands in total. The number of bands per primer ranged from 6-10 as shown in Fig. 2. The genetic distance among the accessions ranged from 0.317073– 0.551724, the closest accessions were 1) control and 3) 12.5Kr of dry

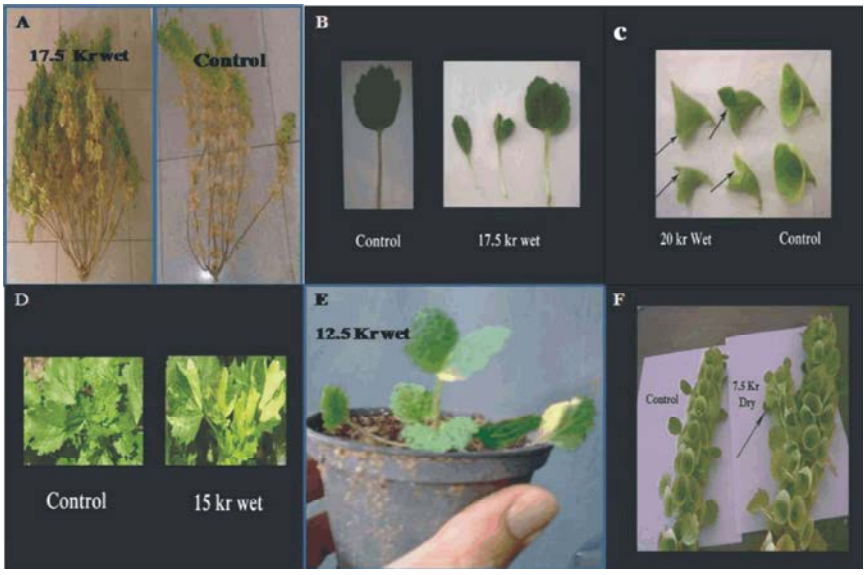

Fig. 1: Showing the plant abnormalities of *Moluccella laevis* as affected by different doses of gamma rays A) The increase of branches number as a resulted of 17.5 Kr of wet seeds, B) leaf abnormalities as a resulted of 7.5 Kr of wet seeds, C) abnormal calyx as a resulted of 20 Kr of wet seeds, D) Variegated leaves as a resulted of 15 Kr of wet seeds, E) Variegated leaves as a resulted of 12.5 Kr of wet seeds, F) abnormal spick as a resulted of 7.5 Kr of dry seeds.

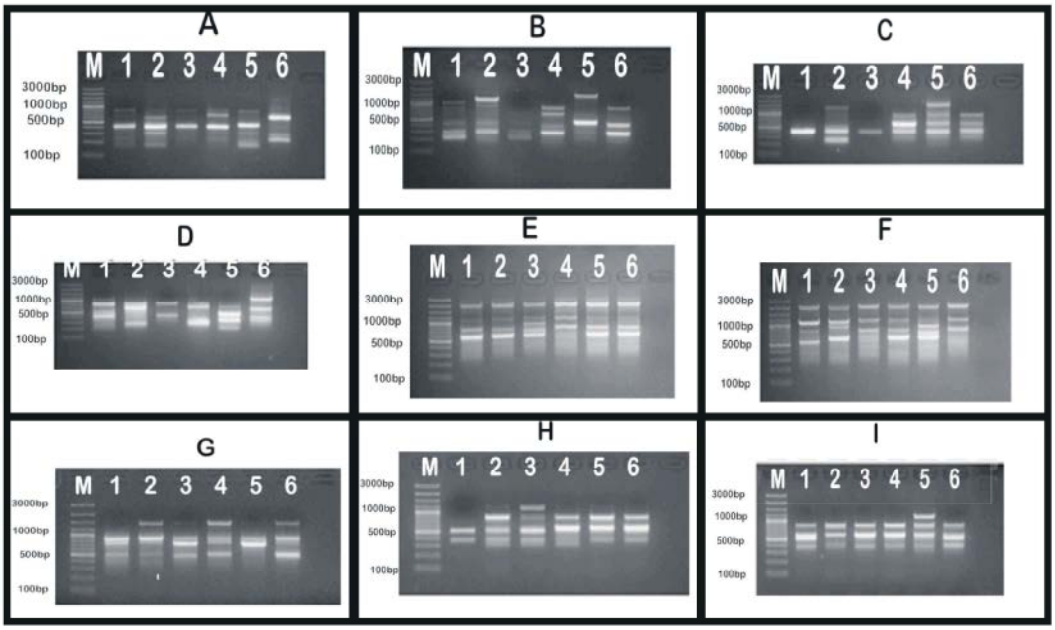

Fig. 2: RAPD profiles of A) OPD 05, B)RAPDA1, C) UBC 162, D) UBC 232, E)RAPDA3, F) RAPDA4,G)RAPDA2, H)RAPDA5 & I)RAPDA6 of six accessions of gamma-irradiated *Moluccella laevis* where 1)Control, 2)15 Kr of wet seeds in the second season, 3)12.5Kr of dry seeds in the first season, 4)12.5Kr of dry in the first season, 5) 17.5 Kr of dry in the first season, 6) 20 Kr of wet seeds in the first season and M)100 bp Molecular weight marker.

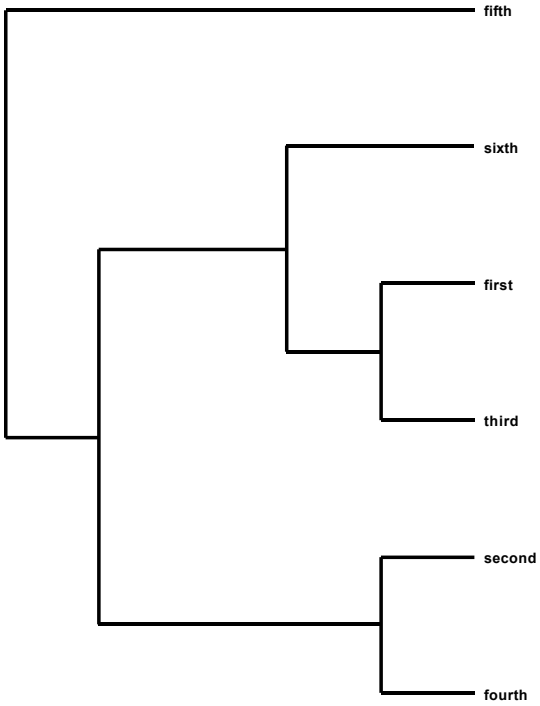

Fig. 3: Dendrogram of six accessions based on UPGMA analysis. The analysis showed the close relatedness between first & third and second and fourth also the analysis showed that the furthest accession was the fifth where: first) Control, second) 15 Kr of wet seeds in the second season, third and fourth) 12.5Kr of dry seeds in the first season, fifth) 17.5 Kr of dry seeds in the first season and sixth) 20 Kr of wet seeds in the first season.

Table 6: Genetic distances based on mean character differences.*

	1	2	3	4	5	6
1	0					
2	0.509804	0				
3	0.317073	0.46	0			
4	0.519231	0.425926	0.470588	0		
5	0.538462	0.551724	0.54717	0.534483	0	
6	0.458333	0.509091	0.5	0.462963	0.45283	0

*Where 1) Control, 2)15 Kr of wet seeds in the second season (Variegated plant), 3)12.5Kr of dry seeds in first season (Leaf shape), 4)12.5Kr of dry seeds in the first season (Flower shape), 5) 17.5 Kr of dry seeds in the first season (Tall plant), 6) 20 Kr of wet seeds in the first season (Dwarf plant)

seeds in the first season and the furthest were 2) 15 Kr of wet seeds in second season and 5)17.5 kr. Dry in the first season. The primers c) UBC162 and D) UBC232 produced the highest number of polymorphic bands (10). The remaining primers showed slightly lower polymorphism and the lowest one was found in G) RAPDA2 (Table 6).

The dendrogram based on genetic distance Unweighted Pair Group Method of Arithmetic Means (UPGMA) (Fig. 3) indicated that the gamma-irradiated *Moluccella laevis* accessions differ significantly in their genetic characteristics also revealed that the furthest accession was Number 5) 17.5 Kr of dry seeds in the first season. The genetic relationship can be useful for designing strategies for breeding programs to produce recombinant hybrid genotypes with superior phytochemical composition and biomass yield [39].

CONCLUSION

The results of the experiments indicated that higher dosage of gamma radiation reduced germination percentage, number of survival plants and plant height. Also, wet treatments of radiation caused a simulative effect in most characters. The high doses 12.5 to 17.5 Kr of wet seeds caused some morphological variations. The genetic relationship of the morphological variations can be determined by using RAPD analysis.

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