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STUDIES ON INDUCING MUTATIONS IN BOUGAINVILLEA

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ABSTRACT: The present work was carried on *Bougainvillea spp* at the Experimental Farm of Agriculture, Zagazig University, during the two successive seasons of 2005 and 2006.The aim of this investigation was to study the response of two cultivars of Bougainvillea; i.e, *B. glabra* (white flowering variety) and *B. buttiana* Mrs Butte (red flowering variety) to physical mutagen gamma ray (0.0, 2.0, 4.0 and 6.0 Krad.) and chemical mutagen (N-nitroso-N methyle Urea) NMU 0.000, .010, 0.015, 0.020 and 0.025% .

Mutations in growth traits, i.e., (compact plants and dwarfness), bracts colour, delay flowering, flower bearing, leaf shape and chlorophyll mutation, flower cluster, length and fasciata were appeared.

In M_2, NMU was efficient than gamma radiation and the variation and the mutations in *B. buttiana* Mrs Butte (red flowering variety) were higher than that of *B. glabra* (white flowering variety).

Key Words: *Inducing Mutations, bougainvillea spp,* physical mutagen gamma ray, (N-nitroso-N methyle Urea).

INTRODUCTION

Bougainvillea (Family Nyctaginaceae) are woody, tropical and subtropical plants, mainly climbers, which excite anyone who seem them in full bloom as they are truly spectacular with their brilliant massed colossus, which last for many varieties make excellent pot plants if their cultural demands are properly understood. Colours of the ornamental cut flowers are governed by both genetic and environmental factors. It has been found that different genes can control various colours of the ornamental plant, (Banerji and Datta, 1987).

Mutations are naturally or artificially induced due to change of the genetic information

contained in the cell. The modern day hybrids of Bougainvillea spectabilis (*B. brasiliensis*) and *B. glabra* are among the most beautiful of flowering vines. The vibrant color of this vine comes not from the small white tubular flowers, but from the 3 large paper like bracts that surround each flower much like the poinsettia.

Many workers studied the effect of gamma irradiation on growth and flowering of floricultural plants. Banerji and Datta (1987), Datta *et al.* (2001) and El Ansary (2003) he found that on cuttings of bougainvillea, gamma rates at 0.0, 0.75 and 1.0 k rad and planted alongside not treated cuttings, and that reductions in plant height, both number of branches and leaves/ plant, leaf size, delayed flowering and number of flowers were noted in treated plants by gamma irradiation.

As for the chemical mutagen; it has been noted that the flowering duration of plants treated with (SA) sodium azide was longer than the flowering duration of those treated with (DES) diethyl sulphate. Finally, it was noted that increasing the concentrations of both mutagens increased the flowering duration more than the control plants in all generations, (Abd ELHady, 2007)

The aim of this work was to induce mutations by the aid of physical mutagen (gamma-rays) and chemical mutagen (N. nitrose- N methyl urea) in Bougainvillea flowers and different shapes or colours of foliage as well.

MATERIALS AND METHODS

The present work was carried out on *Bougainvillea spp* at the Experimental Farm of the Faculty of Agriculture, Zagazig University, at Zagazig, during the two successive seasons of 2005 and 2006. The aim of this investigation was to induce mutations in two cultivars of Bougainvillea; i.e, *B. glabra* white bracts (white flowering variety) and *B. buttiana* Mrs Butte (Red flowering variety). Physical (gamma ray) and chemical (N-Nitroso-N-methyl Urea) NMU mutagens were used for this purpose.

Doses and Concentrations of Mutagens Used

Gamma ray doses

The doses used from gamma rays were 0.0, 2.0, 4.0 and 6.0 Kr. Gamma rays used were generated from Cobalt-60 source, in gamma cell installed in Irradiation Laboratory at Middle East

Regional Radio-Isotope center for Arab country, Cairo, Egypt. The cobalt source emitting radiant energy of 86 rad/second (it's called chronic irradiation).

N-Nitroso-N-methyl urea (NMU) concentrations

The concentrations used from NMU were 0.000, 0.010, 0.015, 0.020, 0.025%. From the stoke solution, freshly prepared solution each concentration was prepared, the NMU used in this study was obtained from Merk W- Germany.

Treated Bougainvillea Cuttings

Treating bougainvillea rooted cuttings by gamma ray

Thirty uniforms cuttings of 20-25cm length of 8-12 mm thickness containing 5-7 buds, chosen from mother plants, were prepared from each Bougainvillea variety for each gamma dose. These cuttings were planted on February 10, 2005, in polyethylene bags (20cm). Filled with mixture of clay, sand and peat moss (1:1:1 by volume, respectively, Mostafa *et al.*, 1996). The cuttings were treated with IBA at 2000 ppm for 15 minutes before planting in the bags. The bags containing cuttings were kept in the nursery in plastic house for 45 days. On April 1, 2005, the rooted cuttings were subjected to gamma ray doses (thirty bags for each dose).

Treating cuttings by NMU

Thirty uniform cuttings of 20-25 length of 8-12 mm thickness containing 5-7 buds chosen from mother plants were prepared for each NMU concentration. These cuttings were soaked in the respective freshly prepared solution for each concentration for 12 hours. Then, they immediately washed in running water to remove excess solution from the cutting surface.

The treated cuttings, after washing, were also soaked in IBA at 2000 ppm for 15 minutes, as in gamma ray. Then, planted in polyethylene bags filled with mixture of clay, sand and peat moss (1:1:1 by volume, respectively; Mostafa *et al.* 1996), then the bags were kept in plastic house.

On April 1, 2005, all the rooted cuttings; i.e. from four gamma rays doses and five NMU concentrations, both varieties were transplanted in the open field was 60 apart and 100cm width. Each treatment were replicated (10 plants each) in a randomized complete block design.

Data Recorded

The following parameters in the M_1 and M_2 (first and second mutanted generation, 2005 and 2006) were recorded after six months. Plant height ,branches number, leaf number, leaf dimensions, flowers number per plant and Leaf chlorophyll (after six months).

Analysis of Chlorophyll a and b (Basic operation of SPAD)

GAL.

Set the power switch to on calibrate meter. Press the measuring head closed without inserting a leaf.

N=O: Take measurement. Insert a leaf and press the measuring head closed.

N= l: 40.0 Up to 30 data automatically stored. (Minolta Co. LTD Japan).

Morphological Changes and Induced Mutations

Plant height (dwarf), growth habit (compact), stem structure (fasciata and branches number), leaf structure (leaf shape) and chlorophyll mutation.

Flowers (bract colour, flowers cluster length, flowers bearing habit, delay flowering and delaless).

Second Mutagenic Generation (M_2-Generation)

Uniform cuttings from every plant in every treatment and replicate in the M_1 generation, has been prepared (20-25cm length, with 8 - 12 mm diameter and 5-7 buds) from the two varieties (white and red). Cuttings were planted as meutiand and before in similar media and in plastic bags, on February 10, 2006. On April 1, 2006, the rooted cuttings were transplanted in the field.

All parameters of M_1 generation were measured in the some manners mentioned in the M_1-generation. Variations in the M_2 - generation included bracts colors.

Statistical Analysis

1- Analysis of variations, (F-test and mean comparison by LSD) for each mutagen, each variety was run according in each generation was run according completely randomized block design (Snedecor and Cochran, 1980).

2- Comparing variation observed and tested it significant; larger mean square/smaller mean square (accord to, Snedecor and Cochran, 1980, A test of equality of two variances.

3- Mutation frequency and spectrum.

RESULTS AND DISCUSSION

Results of physical (gamma ray) and chemical (NMU) mutagens in first (M1) and second (M2) mutanted generations of bougainvillea plants (white var. and red var.) will be presented below. It is known that, the effect of mutagens in M_1- generation was mostly on the plant physiology, but M_2 was the mutanted generation, in a sexual propagated plants.

In a vegetatively propagated plants, assessing the mutations appeared in M_2, and comparing the variation resulted from mutagens treatments and comparing the response of the plant type (white var. and red var.) is useful.

Plant Height

Data in Table 1 revel that treating cuttings with gamma-rays and NMU resulted in a highly significant reduction in M_1-generation and in both bougainvillea types.

However, the reduction in plant height was highly significant with the increase of mutagens treatment in M_1 and M_2 - generations and white and red bougainvillea types.

Comparing the variation mean square Table 2, results indicated insignificant difference between gamma-ray and NMU, between white and red bougainvillea types, and M_1 and M_2-generations.

The results are in accordance with those found by, Banerji *and Datta* (1987) on cuttings of Bougainvillea cv. Roseville and cv. Los Banos Beauty, Datta (1995) on Lantana depressa plants, Baboo *et al.* (1996) on cuttings of Acalypha and El Ansary (2003) on Bougainvillea. They showed that reductions in plant height was noted in treated plants and the effect increased with irradiation dose.

As for the effect of chemical mutagens, Boora *et al.* (2003) on bulbs of tuberose and El Tony (2008) on *Polianthes tuberose* found that the reduction in plant height was increased with increasing the mutagens treatments.

Branch Number

In M_1-generation Table 3, the effect of both mutagens treatment was similar to that at four months, that was true in both Bougainvillea types.

Table 1. Effect of physical (gamma ray) and chemical (NMU) mutagens on plant height (cm) after six months of bougainvillea treated cutting in the M_1 and M_2 - generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
Effect of (gamma ray Kr)				
0 Kr	103.85	101.32	101.10	101.32
2 Kr	100.47	97.85	97.98	98.35
4 Kr	97.83	94.17	95.43	95.27
6 Kr	94.11	91.76	90.75	91.71
F. test.	**	**	**	**
L.S.D at 5%	1.678	3.273	2.822	2.608
$\overline{X}$	99.14	96.27	96.32	96.66
Effect of (NMU, %)				
0.000%	104.18	102.48	103.70	107.06
0.010%	98.64	98.95	101.59	105.90
0.015%	97.75	97.83	99.40	102.02
0.020%	93.99	97.25	97.90	103.99
0.025%	93.63	96.05	96.79	99.93
F. test	**	**	**	**
L.S.D at 5%	2.890	2.367	2.372	0.705
$\overline{X}$	97.64	98.51	99.87	104.38

** = Highly significant

Table 2. Comparison the variation resulted from (gamma ray) and (NMU) mutagen of plant height (cm) after six months of bougainvillea treated cutting in the M_1 and M_2 - generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
M_1.r.g	n.s	M_1.w.c	n.s	M_1.w.g	n.s	M_1.w.c	n.s
M_1.w.g	1.014	M_1.r.c	3.043	M_1.r.c	2.877	M_1.w.g	1.057
M_2.w.g	n.s	M_2.w.c	n.s	M_1.w.c	n.s	M_2.w.g	n.s
M_2.r.g	1.127	M_2.r.c	1.045	M_1.r.g	1.043	M_2.w.c	2.453
M_2.w.g	n.s	M_1.w.c	n.s	M_2.w.g	n.s	M_1.r.g	n.s
M_1.w.g	1.106	M_2.w.c	2.346	M_2.r.c	2.564	M_1.r.c	2.917
M_1.r.g	n.s	M_2.r.c	n.s	M_2.r.g	n.s	M_2.r.g	n.s
M_2.r.g	1.033	M_1.r.c	1.241	M_2.w.c	2.176	M_2.r.c	2.274

n.s. = Not significant
M_1 : First mutanted generation (2005)
M_2: Second mutanted generation (2006)
c: chemical mutagen
w: White colour
r : red colour
g: gamma-ray (α)

Table 3. Effect of physical (gamma ray) and chemical (NMU) mutagens on Number of branches after six months of bougainvillea treated cutting in the M_1 and M_2 - generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
Effect of (gamma ray Kr)				
0 Kr	4.96	5.36	7.22	5.40
2 Kr	5.33	6.17	9.35	6.95
4 Kr	8.29	10.25	8.93	6.95
6 Kr	6.40	7.10	8.25	5.53
F. test.	*	*	**	N.S
L.S.D at 5%	2.045	2.689	1.000	-
$\overline{X}$	6.24	7.22	8.44	6.21
Effect of (NMU, %)				
0.000%	8.31	8.08	8.32	8.98
0.010%	8.21	9.32	8.90	7.79
0.015%	7.82	9.29	8.54	8.66
0.020%	7.17	8.18	8.61	7.97
0.025%	7.58	9.10	8.24	8.75
F. test	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-	-	-
$\overline{X}$	7.82	8.79	8.52	8.43

N.S = Not significant * = Significant ** = Highly significant

In M_2-generation, the effect appeared only with gamma-rays treated white type Bougainvillea, and it was highly significant. NMU, on the other hand, recorded insignificant effect on branches number in M_1, M_2 and both types of Bougainvillea.

Comparing the variability produced mutagens in both types of bougainvillea Table 4, only two cases showed significant high variability, the first one was with NMU treated white type compared with red flowered, and in M_2 red type bougainvillea with NMU had more variability than M_1.

These results agreed with those reported by Youssef *et al.* (2000) on geranium cuttings. They showed that a significant increase in number of branches/ plant occurred when cuttings were exposed to 2 k.rad gamma rays, while a marked decrease was occurred by 4k.rad.

Leaf Number

In M_1 generation Table 5, gamma rays and NMU treatments had insignificant effect on number of leaves /plant of both bougainvillea types. However, highly significant effect on white coloured one with NMU treatment was obtained. So that, the effect of mutagenic treatment in M_1-generation did not extend up to six months. In M_2- generation, highly significant effect on leaf number was observed with gamma-rays and NMU in both bougainvillea types.

Regarding variation induced by gamma rays and NMU Table 6, white coloured bougainvillea reflected higher mean square with gamma rays and NMU in M_1-generation compared with red coloured one treated by NMU. In this respect, M_2-generation had more variation than M_1- generation in red coloured Bougainvillea treated with NMU. In this respect, it could be say that, red coloured bougainvillea had more tolerance to mutagenic treatment, especially in M_1 generation, but gave higher variation in M_2- generation.

El Kholy (1987) on *Hyoscyamus muticus*; Gad (1988) on violet plants and Lamseejan *et al.* (2000) on Chrysanthemum. They found that the number of leaves was increased when rooted cuttings were exposed to doses of gamma rays.

In this regard, Khalaf (2008) exposed dry seeds of *Amaranthus caudatus* to 0, 5, 10, 15 and 20 kr.

Table 4. Comparison the variation resulted from (gamma ray) and (NMU) mutagens on number of branches after six months of bougainvillea treated cutting in the M_1 and M_2 generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
M_1.r.g	n.s	M_1.w.c	*	M_1.w.g	n.s	M_1.w.g	*
M_1.w.g	2.053	M_1.r.c	34.782	M_1.r.c	5.915	M_1.w.c	10.267
M_2.w.g	n.s	M_2.r.c	n.s	M_1.r.g	*	M_2.w.g	*
M_2.r.g	1.172	M_2.w.c	1.381	M_1.w.c	21.081	M_2.w.c	12.820
M_1.w.g	n.s	M_1.w.c	n.s	M_2.w.g	n.s	M_1.r.g	*
M_2.w.g	2.570	M_2.w.c	2.702	M_2.r.c	3.174	M_1.r.c	12.145
M_1.r.g	n.s	M_2.r.c	*	M_2.r.g	*	M_2.r.g	n.s
M_2.r.g	6.187	M_1.r.c	17.776	M_2.w.c	10.936	M_2.r.c	2.606

n.s = Not significant
*= Significant at 0.05%
M_1 : First mutanted generation (2005)
M_2: Second mutanted generation (2006)
w: White colour
g: gamma-ray (α)
r: red colour
c: chemical mutagen

Table 5. Effect of physical (gamma ray) and chemical (NMU) mutagens on number of leaves after six months of bougainvillea treated cutting in the M_1 and M_2 generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
Effect of (gamma ray Kr)				
0 Kr	113.17	105.07	114.88	105.19
2 Kr	114.99	111.53	115.90	112.79
4 Kr	122.07	118.64	120.40	120.54
6 Kr	119.67	122.33	121.36	119.08
F. test.	N.S	N.S	**	**
L.S.D at 5%	-	-	2.818	6.973
$\overline{X}$	117.47	114.39	118.13	114.40
Effect of (NMU, %)				
0.000%	125.65	126.62	122.40	121.64
0.010%	124.61	123.99	121.72	119.13
0.015%	134.54	129.45	125.25	126.11
0.020%	127.78	126.54	122.43	123.19
0.025%	129.11	123.35	118.62	119.92
F. test	**	N.S	**	**
L.S.D at 5%	4.543	-	2.879	2.165
$\overline{X}$	128.34	125.99	122.08	122.00

N.S = Not significant
** = Highly significant

gamma-rays. He noted that the high doses of gamma rays (10, 15 and 20 kr.) caused significant increase in the number of leaves per plant in both generations of both seasons.

With respect to the effect of chemical mutants, Hentrich and Beger (1974) on leaf cuttings of *Saintpaulia ionantha* cultivar Enzett Magdeburger, El Meligy (1981) on corms *of Gladiolus* cv. Eurovision ; Gad (1988) on *Viola plants* and Boora *et al.* (2003) on bulbs of tuberose found that treated plants with chemical mutants recorded high number of leaves/ plant.

In this connection, El Tony (2008) on *Polianthes tuberosa* found that, the chemical mutagen, 0.1% DES increased the number of leaves in the M_1 in both seasons and in M_2 generation of the 2nd season. Generally the increase in SA concentration caused a decrease in the number of leaves in both generations of both seasons. Treatment with 0.1% colchicine for 48hrs. increased the number of leaves. In addition, colchicine was the most effective on increasing the number of leaves per plant.

Leaf Dimensions

Leaf length

Mutagenic treatments of gamma rays and NMU had insignificant effect on leaf length Table 7 when compared with (X) or with mutagen in each, respectively. Moreover, insignificant effect was also observed among doses of each mutagen, i.e., gamma ray or NMU, in M_1 and M_2 generations of mutagens doses.

It is clear from data in Table 8 that comparing mean squares variations of leaf length had insignificant test for all comparisons.

Leaf width

Mutagenic treatments in M_1 and M_2 generations in both Bougainvillea types, had insignificant variation in M_1 and M_2- generations and both bougainvillea types Tables 9 and 10, when comparing mean squares results for mutagenic treatments.

Flower Number per Plant

Results of flower number per plant Tables 11 and 12 show significant response to gamma rays in M_1 only in both bougainvillea types. While N M U had no

Table 6. Comparison the variation resulted from (gamma ray) and chemical (NMU) mutagens on number of leaves after six months of bougainvillea treated cutting in the M_1 and M_2 generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
M_1.r.g	n.s	M_1.w.c	*	M_1.w.g	*	M_1.w.g	n.s
M_1.w.g	3.484	M_1.r.c	34.782	M_1.r.c	38.765	M_1.w.c	1.114
M_2.r.g	n.s	M_2.r.c	n.s	M_1.r.g	n.s	M_2.w.g	n.s
M_2.w.g	4.815	M_2.w.c	1.381	M_1.w.c	3.883	M_2.w.c	1.855
M_1.w.g	n.s	M_1.w.c	n.s	M_2.w.g	n.s	M_1.r.g	*
M_2.w.g	1.623	M_2.w.c	2.702	M_2.r.c	1.343	M_1.r.c	135.06
M_1.r.g	n.s	M_2.r.c	*	M_2.r.g	n.s	M_2.r.g	n.s
M_2.r.g	1.198	M_1.r.c	17.776	M_2.w.c	8.756	M_2.r.c	6.339

n.s. = Not significant
*= Significant
M_1 : First mutanted generation (2005)
M_2: Second mutanted generation (2006)
w: White colour
r : red colour
g: gamma-ray (α)
c: chemical mutagen

Table 7. Effect of physical (gamma ray) and chemical (NMU) mutagens on leaf dimensions (leaf length) after six months of bougainvillea treated cutting in the M_1 and M_2- generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
	Effect of (gamma ray Kr)			
0 Kr	4.32	4.21	4.23	3.77
2 Kr	4.28	4.14	4.51	3.42
4 Kr	4.19	3.68	4.50	3.51
6 Kr	4.54	4.14	4.48	4.03
F. test.	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-	-	-
$\overline{X}$	4.33	4.04	4.43	3.68
	Effect of (NMU, %)			
0.000%	4.01	4.20	4.30	3.87
0.010%	3.69	3.76	4.02	3.65
0.015%	4.03	3.43	4.03	3.35
0.020%	4.09	3.36	3.00	3.71
0.025%	3.74	3.60	4.21	3.65
F. test	N.S	*	N.S	N.S
L.S.D at 5%	-	0.507	-	-
$\overline{X}$	3.91	3.67	3.91	3.64

N.S = Not significant * = Significant

Table 8. Comparison the variation resulted from (gamma ray) and chemical (NMU) mutagens on leaf dimensions (leaf length) after six months of bougainvillea treated cutting in the M_1 and M_2- generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
M_1.r.g	n.s	M_1.r.c	n.s	M_1.r.c	n.s	M_1.r.c	n.s
M_1.w.g	2.573	M_1.w.c	3.132	M_1.w.g	4.882	M_1.w.g	4.882
M_2.r.g	n.s	M_2.r.c	n.s	M_1.r.g	n.s	M_1.r.g	n.s
M_2.w.g	4.264	M_2.w.c	1.945	M_1.w.c	1.650	M_1.w.c	1.650
M_1.w.g	n.s	M_1.w.c	n.s	M_2.r.c	n.s	M_2.r.c	n.s
M_2.w.g	1.283	M_2.w.c	1.927	M_2.w.g	2.018	M_1.w.g	2.018
M_2.r.g	n.s	M_1.r.c	n.s	M_2.r.g	n.s	M_2.r.g	n.s
M_1.r.g	1.291	M_2.r.c	3.112	M_2.w.c	4.109	M_2.w.c	4.109

n.s = Not significant
M_1 : First mutanted generation (2005)
M_2: Second mutanted generation (2006)
w: White colour
r : red colour
g: gamma-ray (α)
c: chemical mutagen

Table 9. Effect of physical (gamma ray) and chemical (NMU) mutagens on leaf of dimensions (leaf width) of bougainvillea treated cutting in the M_1 and M_2- generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
	Effect of (gamma ray Kr)			
0 Kr	3.29	3.20	3.01	3.08
2 Kr	3.36	3.18	2.78	2.63
4 Kr	3.05	2.79	3.15	2.57
6 Kr	2.94	2.83	3.07	3.10
F. test.	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-	-	-
$\overline{X}$	3.16	3.00	3.00	2.84
	Effect of (NMU, %)			
0.000%	2.84	3.10	2.54	3.06
0.010%	2.80	2.88	2.47	2.86
0.015%	2.44	2.54	2.55	2.50
0.020%	2.65	2.52	2.59	2.46
0.025%	2.59	2.81	2.80	2.76
F. test	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-	-	-
$\overline{X}$	2.66	2.77	2.59	2.72

N.S = Not significant

Table 10. Comparison the variation resulted from (gamma ray) and Chemical (NMU) on leaf dimensions (leaf width) after six months of bougainvillea treated cutting in the M_1 and M_2 generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
M_1.r.g	n.s	M_1.r.c	n.s	M_1.w.c	n.s	M_1.w.g	n.s
M_1.w.g	1.218	M_1.w.c	2.256	M_1.w.g	1.558	M_2.w.c	1.236
M_2.r.g	n.s	M_2.r.c	n.s	M_2.w.c	n.s	M_2.w.c	n.s
M_2.w.g	3.155	M_2.w.c	3.938	M_2.w.g	1.037	M_2.w.g	1.037
M_1.w.g	n.s	M_1.w.c	n.s	M_1.r.c	n.s	M_1.r.c	n.s
M_2.w.g	1.545	M_2.w.c	1.591	M_1.r.g	1.897	M_1.r.g	1.897
M_2.r.g	n.s	M_2.r.c	n.s	M_2.r.g	n.s	M_2.r.g	n.s
M_1.r.g	1.675	M_1.r.c	1.096	M_2.r.c	2.112	M_2.r.c	2.112

n.s = Not significant

M_1 : First mutanted generation (2005)

M_2: Second mutanted generation (2006)

w: White colour

r : red colour

g: gamma-ray (α)

c: chemical mutagen

Table 11. Effect of physical (gamma ray) and chemical (NMU) mutagens on number of flowers after six months of bougainvillea treated cutting in the M_1 and M_2-generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
Effect of (gamma ray Kr)				
0 Kr	54.90	65.30	60.03	89.03
2 Kr	55.24	64.27	60.35	82.76
4 Kr	61.53	71.43	63.62	83.97
6 Kr	56.47	66.09	61.26	84.71
F. test.	**	*	N.S	N.S
L.S.D at 5%	3.079	4.291	-	-
$\overline{X}$	57.03	66.77	61.31	85.12
Effect of (NMU, %)				
0.000%	71.56	85.67	81.04	100.86
0.010%	75.56	84.81	76.86	99.98
0.015%	71.32	85.70	84.22	100..93
0.020%	74.32	85.74	77.23	99.52
0.025%	74.45	85.09	77.82	99.31
F. test	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-		-
$\overline{X}$	74.04	85.40	79.43	100.12

N.S = Not significant * = Significant ** = Highly significant

Table 12. Comparison the variation resulted from (gamma ray) and (NMU) mutagens on number of flowers after six months of bougainvillea treated cutting in the M_1 and M_2 generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
$\frac{M_1.r.g}{M_1.w.g}$	n.s 1.079	$\frac{M_1.r.c}{M_1.w.c}$	n.s 2.621	$\frac{M_1.w.g}{M_1.r.c}$	n.s 3.901	$\frac{M_1.w.g}{M_1.w.c}$	n.s 4.303
$\frac{M_2.r.g}{M_2.w.g}$	n.s 2.829	$\frac{M_2.w.c}{M_2.r.c}$	n.s 3.504	$\frac{M_1.r.g}{M_1.w.c}$	n.s 4.584	$\frac{M_2.w.g}{M_2.w.c}$	n.s 1.338
$\frac{M_1.w.g}{M_2.w.g}$	n.s 3.587	$\frac{M_1.w.c}{M_2.w.c}$	n.s 1.115	$\frac{M_2.w.g}{M_2.r.c}$	n.s 4.690	$\frac{M_1.r.g}{M_1.r.c}$	n.s 1.772
$\frac{M_1.r.g}{M_2.r.g}$	n.s 1.369	$\frac{M_1.r.c}{M_2.r.c}$	* 10.250	$\frac{M_2.r.g}{M_2.w.c}$	n.s 3.786	$\frac{M_2.r.g}{M_2.r.c}$	* 13.269

n.s = Not significant
M_1 : First mutanted generation (2005)
M_2: Second mutanted generation (2006)
w: White colour
r : red colour
g: gamma-ray (α)
c: chemical mutagen

significant effect on flower number in M_1 and M_2, generations in both bougainvillea types.

These results agreed with those obtained by Kannan *et al.* (2002) who studied the effect of gamma rays at 1.5 and 2.0 kR, EMS at 30 and 45 mM and 1.0 kR+15mM and 1.5kR+30 mM of combined treatments on floral characters of *Jasminum sambac* cv. Gundumalli. They indicated that the mean values of the reproductive traits; namely, length of the corolla tube, length of flower bud, width of the flower bud and diameter of the open flower after mutagenesis showed, in general, positive shifts over control population in all doses except the higher ones. Also, Abd El Hamed (2004) found that, on narcissus plants, the highest increment in the number of flowers during the two seasons was obtained at 5 or 7.5 kr of gamma rays.

Leaf Chlorophyll

In both M_1 and M_2 generations Table 13, gamma rays and NMU treatments had insignificant effect on leaves chlorophyll content in both bougainvillea types.

Comparing variation Table 14, significant mean squares (of red flowered one) resulted from gamma ray was observed when compared with mean squares due gamma ray in M_1 and M_2 generations. Such variation, declared that NMU produced variations in M_1 generation in both red and white flowers of bougainvillea. It was observed that NMU showed higher variation in M_2 generation in both bougainvillea types.

Observations in the M_2 Generation

Mutations Characteristics

Effect of Gamma Rays on White Variety

Growth habit (compact plant)

Data in Tables 15 and 16 show that the treatment of 6 kr gamma ray caused compact plant (18.75%).

These results agreed with those reported by Park and Anderson (1990), they observed irradiated of *Passiflora caerulea* cuttings with gamma rays at doses of 15-60 Gy changed in growth habit.

A range of gamma rays from 10 to 60 Gy was applied to apical and nodal microcuttings of five species of *Lonicera*. From 1700 plants produced from irradiated buds, only those from *L. nitida* Maigrun

Table 13. Effect of physical (gamma ray) and chemical (NMU) mutagens on leaf chlorophyll content of bougainvillea treated cutting in the M_1 and M_2- generations (2005 and 2006)

Treatments	M_1		M_2	
	White variety	Red variety	White variety	Red variety
	Effect of (gamma ray Kr)			
0 Kr	32.84	37.57	30.31	36.95
2 Kr	31.42	37.44	34.21	40.51
4 Kr	32.73	36.47	36.66	43.26
6 Kr	30.92	38.59	35.50	43.98
F. test.	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-	-	-
$\overline{X}$	31.977	37.517	34.42	41.175
	Effect of (NMU, %)			
0.000%	32.32	40.12	34.51	39.65
0.010%	30.24	35.41	33.84	39.48
0.015%	31.34	35.69	33.83	39.39
0.020%	31.37	41.71	32.16	44.66
0.025%	30.43	39.22	32.75	51.23
F. test	N.S	N.S	N.S	N.S
L.S.D at 5%	-	-	-	-
$\overline{X}$	31.14	38.43	33.418	42.882

N.S = Not significant

Table 14. Comparison the variation resulted from (gamma ray) and (NMU) mutagen of leaf chlorophyll content of bougainvillea treated cutting in the M_1 and M_2 - generations (2005 and 2006)

Mutagen				Comparison			
Gamma		Chemical		Gamma		Chemical	
M_1.w.g / M_1.r.g	n.s 1.208	M_1.r.c / M_1.w.c	* 10.48	M_1.r.c / M_1.w.g	n.s 8.468	M_1.w.g / M_1.w.c	n.s 1.238
M_2.r.g / M_2.w.g	n.s 1.333	M_2.r.c / M_2.w.c	* 30.074	M_1.r.g / M_1.w.c	n.s 1.024	M_2.w.g / M_2.w.c	n.s 8.569
M_2.w.g / M_1.w.g	n.s 8.362	M_2.w.c / M_1.w.c	n.s 1.208	M_2.r.c / M_2.w.g	n.s 3.509	M_1.r.c / M_1.r.g	n.s 10.234
M_2.r.g / M_1.r.g	* 13.471	M_2.r.c / M_1.r.c	n.s 3.465	M_2.r.g / M_2.w.c	n.s 11.423	M_2.r.c / M_2.r.g	n.s 2.633

n.s = Not significant
M_1 : First mutanted generation (2005)
M_2: Second mutanted generation (2006)
w: White colour
r : red colour
g: gamma-ray (α)
c: chemical mutagen

Table 15. Number of survived plants and mutations frequency and percentage of Bougainvillea plant as affected by physical (gamma ray) and chemical (NMU) mutagens in the M_1 and M_2 generations

Treatments	M_1				M_2			
	No. plants White Var.	%	No. plants Red Var.	%	No. plants White Var.	%	No. lants Red Var.	%
Effect of (gamma ray. Kr)								
0 kr	29 0	0	29	0	27 0	0	27 0	0
2kr	25 0	0	25	0	22 (3)	13.63	23 (3)	13.04
4kr	22 0	0	22 (1)	4.54	19 (2)	10.52	19 (2)	10.52
6kr	19 (2)	10.52	19 (1)	5.26	16 (2)	18.75	14 (3)	21.42
average	0.66	3.50	0.66	3.26	2.33	14.3	2.66	14.99
Effect of (NMU%)								
0.000%	28 0	0	29 0	0	27 0	0	28 0	0
0.010%	26 0	0	27 (1)	3.7	25 (1)	4.00	24 (2)	8.33
0.015%	25 0	0	24 0	0	25 (1)	4.00	23 (3)	13.04
0.020%	24 (2)	8.33	24 0	0	22 (2)	9.09	22 (2)	9.09
0.025%	23 0	0	23 (1)	4.34	20 (2)	10.00	22 (3)	13.63
average	0.5	3.12	0.5	2.01	1.50	6.77	2.75	12.16

( ) Number of mutated plants.

Table 16. Spectrum of morphological mutations in different varieties of Bougainvillea plant in M_2 generation

Mutant type	Gamma ray (kr)		NMU (%)		Total average %
	Var. white	Var. red	Var. white	Var. red	
1	2	3	4	5	6
Plant Height					
Dwarf	-	1 (7.14)	-	-	1 (0.032)
Growth habit					
Compact	2 (18.75)	1 (7.14)	1 (4.545)	3 (13.434)	7 (0.22)
Stem structure					
Fasciata	-	-	-	1 (4.543)	1 (0.03)
Branches No	-	-		1 (4.165)	1 (0.032)
Leaf structure					
Leaf shape	-	-	2 (9.545)	1 (4.543)	3 (0.096)
Chlorophyll mutation	1 (5.26)	-	1 (4.00)	-	2 (0.064)
Flowers					
Bract colour	1 (4.54)	3 (14.866)	1 (5.00)	1 (4.346)	5 (0.193)
Flowers cluster length			1 (4.00)	1 (4.346)	2 (0.064)
Flowers bearing Habit	-	3 (15.832)	-	2 (8.71)	5 (0.161)
Delay Flowering					
Delaless	3 (14.346)	-	-	-	3 (0.096)
No. of mutations	7	8	6	10	31
Total No. of mutation types	4	4	5	6	19

were observed during two years. Among 200 of these plants, some compact and slender mutants were detected, (Cambacedes *et al.*, 1992).

Leaf structure (chlorophyll mutation)

The treatment of 4 kr gamma ray caused mutation in chlorophyll mutation rate (5.26 %) in Tables 15 and 16 of plant top, faded colour.

Flowers (bract colour)

The treatment of 2 kr gamma ray induced mutation in the bract colour. It was changed from white to red (4.54 %) in Tables 15 and 16 These results are in line with those reported by Banerji and Datta (1987) exposed *Bougainvillea glabra* Los Banos Beauty cuttings to 0, 0.75 and 1 Krad. Somatic mutation in bract colour was noticed in one branch in chimeric form after treatment with 0.75 Krad. The original bract colour was rhodamine purple, while the mutant bract was rhodamine red. Also rooted cuttings of bougainvillea spectabilis were gamma irradiated with doses of 3 and 3.5 Krad. M_1 mutants with different bract colours were produced. At 3 Krad, mutants with scarlet and orange-yellow bracts were obtained, while at 3.5 Krad mutants with orange-yellow and red-yellow bracts were produced, (Deng and Shaode, 1990).

Delay flowering

The treatment of 2 and 4 kr gamma ray caused delaying in flowering (14.346 %) in Tables 15 and 16.

These results are in accordance with those reported by Lata (1980) who found that irradiated bud-wood of seven rose cultivars with the doses of 0, 3, 4 and 5 Krad from Co 60 source. Delay of 4-6 weeks in the commencement of flowering was apparent in the 4 and 5 Krad treated plants. Long intermittent breaks in the flowering period were commonly observed. A continuous flowering was observed in Pink Parafait. The decrease in blooms correlated with the increase in exposure.

Effect of Gamma Rays on Red Variety

Plant height (dwarfness)

It is clear from data in Tables 15 and 16 that the treatment of 6 kr gamma ray caused plant dwarfness (7.14 %). These results are in harmony with those obtained by Khalaburdin (1993) who studied

the effect of gamma rays at 1, 5, 10 and 15kr on hybrid Canna. It was found that the doses of 15kr. gave the highest percentage of morphological changes including dwarfness. Also, El-Ansary (2003), on Bougainvillea, found that treatments of 2and 3 Krad caused several changes in leaf shape. The treatments of 1, 2 and 3 Krad caused also some changes in leaf shape. Meanwhile, treatments of 2 and 3 Krad caused hard dwarfing of some plants of the cultivar Pink Pixie while treatment of 3 Krad caused a giant mutant of one plant of the cultivar Scarlet O' Hara.

Growth habit (compact plant)

The treatment of 6 kr gamma ray caused compact plant (7.14 %) in Tables 15 and 16.

Flowers (bract colour)

The treatment of 2 and 4 kr gamma ray induced mutations in the bract colour was changed from red to orange-yellow flowers were produced mutation rate 14.866% in Tables 15 and 16.

Flower bearing

The treatment of 2 and 6 kr gamma ray caused in flowers bearing singly on branch compared with control. Also flowers bearing in group on branches top 15.832 % in Tables 15 and 16. The results are in harmony with those found by Deng and Liu (1990) on *Bougainvillea spectabilis.*

Effect of NMU on White Variety

Growth habit (compact plant)

The treatment of 0.020 % NMU caused compact plant 4.545 % in Tables 15 and 16.

Leaf structure (leaf shape)

The treatment of 0.020 and 0.025% NMU caused in change leaf shape 9.545 % in Tables 15 and 16. These results agreed with those reported by El Fadaly (2003) who found that the most effective treatment for increasing the leaves mutation was 1.0 % EMS+ 10 kr. He also found that five mutants in the leaf shape were identified in Catharanthus roseus, while 2x10-3m NaN3+1.0 % EMS gave 15 mutants in the leaf shape of Dimorphotica ecklonis.

Chlorophyll mutation

The treatment of 0.015% NMU caused mutation in chlorophyll mutation rate (4.00%) in Tables 15 and 16.

Flower structure (flower cluster length)

The treatment of 0.010 % NMU caused reduction in flower cluster length compared with control (4.00%) as recorded in Tables 15 and 16.

These results are in accordance with those reported by Warfield (1973) who reported that higher EMS concentrations of 0.25 and 0.50 M to the petioles of *Saintpaulia ionantha* leaf cutting produced mutation. He mentioned that the 0.50 M treatment for 1 h. gave double to single flower. The most extreme treatment was 0.50 M for 1 h. produced double to single flower, extremely early bloom, flecked speckled and abnormal petals mutations. Also, El Tony (2008) on *Polianthes tuberose* found that, in both seasons, there were morphological changes in the plants according to the different doses and concentrations of the mutagenic agents. Changes in leaf shape, compact plant, inflorescence character, spike shape and bulbs size, shape, colour and number were observed.

Effect of NMU on Red Variety

Growth habit (compact plant)

The treatment of 0.010, 0.020 and 0.025% NMU caused compact plant compared with control (13.434%) as shown in Tables 15 and 16.

Leaf Structure (Fasciata and Branches)

Fasciata

The treatment of 0.025% NMU appeared like a cluster of closely fused branches as a result of fasciations (4.543 %) in Tables 15 and 16.

Branches

The treatment of 0.010% NMU caused in increasing number of branches with compared to control (4.165%) in Tables 15 and 16.

Leaf structure (leaf shape)

The treatment of 0.025% NMU caused in change leaf shape (4.346%) in Tables 15 and 16.

Flower Structure (Bract Colour, Flowers Cluster Length and Flower Bearing)

Bract colour

The treatment of 0.015 % NMU caused in mutation in bract colour were changed from red to light red flowers were produced (mutation rate 4.346 %) as shown in Tables 15 and 16.

Flowers cluster length

The treatment of 0.015 % NMU caused in reduction flowers cluster length with compared control (4.346 %), as recorded in Tables 15 and 16.

Flower bearing

The treatments of 0.010 and 0.020% NMU caused in reduced changed flower bearing with compared control (8.74%) as shown in Tables 15 and 16.

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دراســـــات علـــى استحداث طفـرات فـــى الجهنميـــة

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أجريت هذه الدراسة علي نبات الجهنمية في مزرعة كلية الزراعة التجريبية، جامعة الزقازيق خلال موسمي ٢٠٠٥، ٢٠٠٦. هدف هذه التجربة هو دراسة استجابة صنفين من الجهنمية، هما الجهنمية جلابرا (الصنف ذو اللون الأبيض) والجهنمية دم الغزال (الصنف ذو اللون الأحمر) لأشعة جاما (صفر ، ٢,٠، ٤,٠، ٦,٠ كيلو راد) والنتروزوميثيل يوريا (صفر ،٠,٠١٠، ٠,٠١٥، ٠,٠٢٠، ٠,٠٢٥%).

ظهرت الطفرات في صفات النمو الخضري مثل (نباتات مندمجة ومتقزمة)، ولون القنابات، وتأخير الإزهار، وتبكير التزهير، وشكل الورقة، وطفرات كلوروفيلية، وطول الحامل الزهري، وتفريع القمة النامية.

كان تأثير المطفر الكيماوي النيتروزوميثيل يوريا أكثر تأثيرا مقارنة بأشعة جاما، والاختلافات والطفرات كانت أكثر ظهورا في الجهنمية دم الغزال (الصنف ذو اللون الأحمر) عنها في الجهنمية الجلابرا (الصنف ذو اللون الأبيض) وذلك في الجيل الطفري الثاني.