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INDUCTION OF PROMISING MUTANT LINES OF CANOLA UNDER NEW RECLAMATION LANDS (HARSH AND SALINE)

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ABSTRACT

The present study aimed to induce of promising mutants lines of Canola possess high seed yield and oil content under new reclamation desert land at Ras-Suder-Sina (saline) and Inshas (harsh and poor fertility land). Canola seeds of four varieties (Serow4, Serow6, Bactol as local and Evita as exotic varieties) were treated with gamma ray at four doses (0, 100, 400 and 600 Gy). Thirty mutants for number of pods/plant and changes in morphological criteria were selected at M2 generation. The mutants at M3 generation confirmed that induction of mutant lines possessed high number of pods and seed yield/plant than the mother varieties. These mutant lines possessed homogeneity at M3 generation, 8, 11, 18 and 22 mutant lines at serow4, 38 and 45 at serow6, 63 and 66 at Bactol and mutant line 74 at Evita. Highest number of pods/plant (110) was recorded at line 74 from Evita variety. The results were appeared the same trend for seed yield/plant with number of pods/plant, the lines which possessed high number of pods/plant were had high seed yield/plant. The results at M4 and M5 generations for 13 homogeneity mutant lines selected from M3 generation contained different response of mutant genotypes for different conditions on the bases of number of pods and seed yield/plant. Promising mutant lines were detected under both conditions possessed significant at both M4 and M5 generations. These results confirmed that the genetic control of adverse environmental conditions was different between them and probable polygene mechanisms play a role in the inheritance of saline and harsh conditions. Oil percent as well as earliness criterion at M4 and M5 were recorded. Finally six mutant lines possessed promising performance of these study, lines 11 and 66 under both conditions (Suder and Inshas), lines 38 and 63 under Ras-Sudr and lines 74 and 75 under Inshas conditions.

Keywords: Canola, mutant, gamma ray, number of pods / plant, seed yield /plant, oil percent.

INTRODUCTION

Rapeseed and canola are closely related members of the mustard family (Brassicaceae) that are both grown as oil seed crops. All current varieties of rapeseed and canola were developed from *Brassica napus* and *Brassica rapa.*

Rapeseed is grown primarily as a source of erucic acid, which is not edible but is valuable in high-performance industrial lubricants. In the early 1970s, Canadian plant breeders used conventional breeding techniques to remove the anti-nutritional erucic acid and bitter glucosinolates from rapeseed. Removing these compounds resulted in an oil seed crop that produced edible oil low in saturated fats and a very palatable, high-protein meal for animal feed. They coined the word Canola (for Canadian oil low acid) to describe a crop that is low in both compounds. Canola oil is used mainly as cooking oil and in shortening and margarine. To be considered canola, the oil and meal must both meet the following standard: oil < 2% erucic acid, meal < 30 micromoles of glucosinolates per gram.

Because canola oil is very high in unsaturated fatty acids, it is considered a high-quality food oil that is healthy in human diets.

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Canola oil is particularly desirable for frying because it has a neutral flavor and can be heated to higher temperatures than many other oils without smoking or burning.

Canola meal is a high-protein (34-38%) animal feed used by the dairy, cattle, and poultry industries (Ehrensing, 2008).

Availability of genetic variability is the prerequisite for any breeding programme.

Besides conventional methods, induced mutation has been extensively used for creating new genetic variation in crop plants. Literature revealed that more than 2200 mutant varieties of different crops with improved agronomic traits have been developed and released to the farmers for general cultivation all over the world (Maluszynski *et al.*, 2000). Mutagenesis technique has also been successfully employed in rapeseed and mustard by the plant breeders (Naz and Islam 1979 and Javed *et al.*, 2000) to alter the genetic architecture of plant and isolate the possible mutants with desired economic plant characters (Rehman *et al.*, 1987; Robbelen, 1990 and Mahla *et al.*, 1990).

Egypt, like Pakistan, is deficient in edible oil and is continuously meeting the domestic requirement at the cost of precious foreign exchange. In Pakistan during 2006-07, 59.506 billion rupees were spent on the import of 1.787 million tons of edible oil. At present, edible oil requirement of the country is 2.764 million tons annually, of which 0.857 million tons (31%) comes from local resources and 1.907 million tons (69%) is being imported (Anonymous, 2006). Canola (canadian oil, low in acid) is now the third largest source of edible oil after soybean (*Glycine max*) and palm (*Elaeis oleifera*) oil (Nowlin, 1991). Canola is recent introduction in Pakistan and area under this crop is expanding rapidly especially under moderate climatic conditions. However yield is less than potential of existing cultivars due to many reasons, shortage of water being the most important one. Canola is relatively poorly adapted to drought condition (Wright *et al.*, 1997).

In Egypt, one of the most important problems in the agricultural sector is the shortage of oil production, which estimated approximately by 90% of the oil consumption and leads to large amounts importation from abroad. One of the governmental solutions to face such problem is expanding the cultivate area by oil crops or introduction of new oil seed crops like canola. But the limited area for agriculture in old lands, especially at intensive competition between wheat and clover at winter season, any new crops in Egyptian agriculture should be grown under new reclamation desert lands. Therefore, the present study aimed to induction of promising mutant lines of Canola (oil crop) possess high seed yield, and oil content under new reclamation desert lands at Ras Sudr, Sina (saline) and Inshas (harsh and poor fertility land).

MATERIALS AND METHODS

Plant Materials

Four canola varieties (Three local and one exotic) used in this investigation. Serow4, Serow6 and Bactol as local varieties obtained from Oil Crops Res. Dept., Field Crops Res. Inst., ARC, Egypt. Evita as exotic variety introduced from Germany.

Irradiation Treatments

Air-dried seeds of four varieties were exposed to 0, 100, 400 and 600 Gy of gamma rays (C^{60}) at Nuclear res. Center, Inshas, Egypt. The dose rate was 5.6 Gy/minute.

Field Experiments

M_1 generation for four varieties with four treatments (0, 100, 400, 600) Gy of gamma rays were grown at 2005/2006 in Inshas exp, Nuclear res. Center, for obtaining of M_2 seeds. The M_2 generation was grown at 2006/2007 and examination of each M_2 plants for selection of any changes at morphological criteria as well as seed yield and it's attribute, especially number of pods / plant. The M_2 mutants in each variety and treatments were collected and examined. The mutants which possessed highly significant for number of pods and seed yield recorded at Table 1.

In 2007/2008 season , each M3 seeds for each M_2 mutant were grown in three rows with three replication for obtaining M3 generation, in addition the control of each variety. The number of pods/plant and seed yield were recorded for using them in selection and determined the M4 mutant lines.

Table 1. The number of M2 mutants isolated from each variety

Treatment / Varity	0 Gy	100 Gy	400 Gy	600 Gy	Total
Serow 4	-	6	2	2	10
Serow 6	-	1	1	1	3
Bactol	-	3	3	2	8
Evita	-	2	3	4	9
Total	-	12	9	9	30

In 2008/2009 season, the seeds of selected families were sown with their parents to evaluate individually plant progenies (M4) in two locations: Ras Sudr (salty soil) and Inshas (sandy soil), Egypt.

In 2009/2010 season, mutant lines were sown with four parents at two locations, the split plot design with three replications was used during the evaluation two seasons (2008/2009 and 2009/2010). The main plots were assessed to variety and the split plots were used for progenies of selected plants.

LSD was used to compare the mean values according to (Gomez and Gomez, 1983) and the data were collected on the following characters: number of pods / plant, Seed yield (g) /plant, oil percent of seeds and acid value of oil in M4 and M5 generations.

RESULTS AND DISCUSSION

Number of Pods/Plant

High variations were recorded at M3 generation for number of pods /plant (Fig. 1). The range between average mean of number of pods per plant for mother variety were narrow (55-60 pods/plant). In contrast the range between induced mutants within each variety or between over all varieties were wide (55-110 pods/plant). These results confirmed that the success of γ-rays for induction of genetic variability in No. of pods /plant in canola varieties. Four mutant lines (lines 8, 11, 18 and 22) were possessed high number of pods/plant (about 95 pods) in comparison with the mother variety Serow4. While, two mutants only (line 38, 45) were higher than mother variety Serow6, only two mutant lines (line 63, 66) were higher than mother variety (Bactol). Highest number of pods than the almost mutant lines and varieties was recorded at line 74 from Evita variety, which gave 110 pods /plant.

Seed Yield/Plant

Average mean of mutant lines for seed yield(g)/plant were recorded (Fig. 2). These results appeared the same trend of mutant lines. The lines which possessed high number of pods/plant, it had also high seed yield(g)/plant, but the variation between mutant lines for seed yield /plant was lowest than number of pods/plant (3.5-6 g). Line 8 and 66 had higher seed yield than almost mutant lines and varieties. These results agree with (Syed and Rahman, 2009), they mentioned that, all the three varieties possessed high yield potential, medium-to-high oil content, early maturity and broader adaptability to rainfed and irrigated environments in comparison with the local check varieties and respective parents. These varieties are being cultivated by growers on appreciable areas.

According to the homogeneity of mutants and segregation within each mutant line, the mutant lines were rest and continuous to M4 and M5 generations, 13 promising mutant lines at M4 and M5 were selected from 30 mutant lines at M3 generation. The M4 seeds of each mutant and each variety were divided into two parts, the first part sowing at Ras–Sudr–Sina and the other sown at Inshas (harsh land). For obtaining of M4 generation significant and highly significant differences were recorded in M4 and M5 generations at two different environmental conditions (Table 2). Number of pods/plant and seed yield/plant and subsequently high heritability

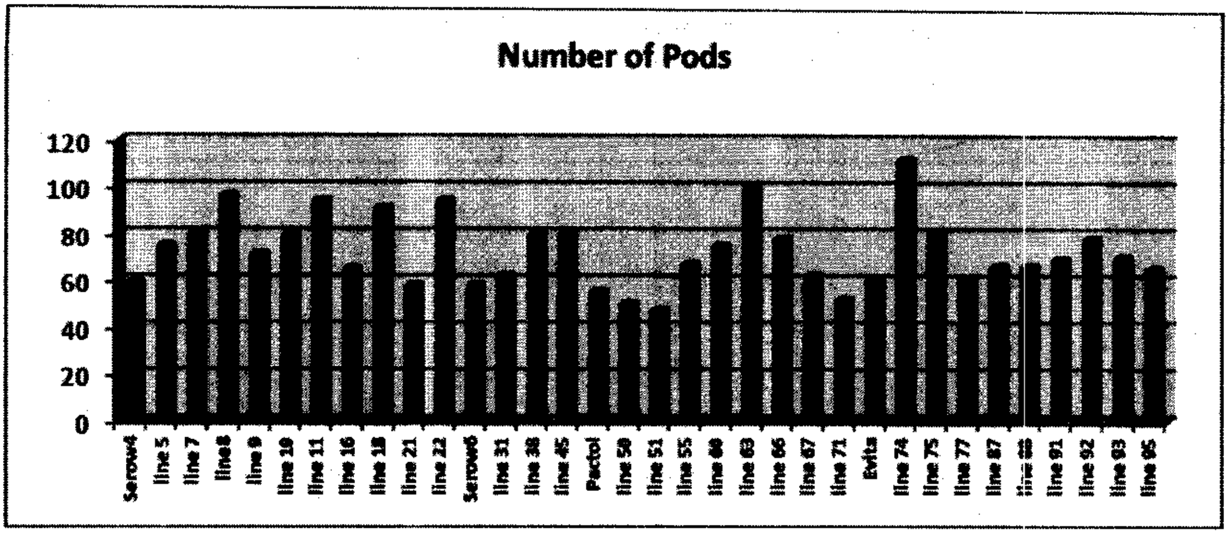

Fig. 1. Average mean of M3 progenies for number of pods /plant for four varieties under study

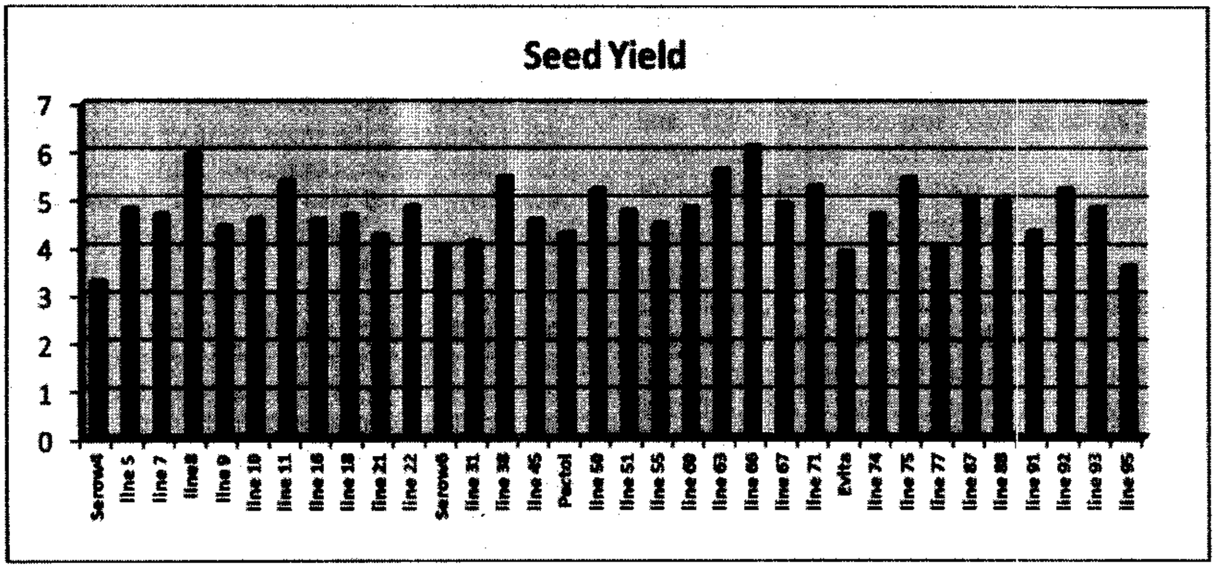

Fig. 2. Average mean of M3 progenies for seed yield /plant for four varieties under study

Table 2. Mean sum of square (MS) and heritability (h^2) for number of pods / plant and seed yield (g)/plant at M4 andM5 generations

		MS		MS		MS		MS	
		No. of pods/Ras-Sudr		Seed yield/ Ras-Sudr		No. of pods/Inshas		Seed yield/Inshas	
Source	**d.f**	M4	M5	M4	M5	M4	M5	M4	M5
Replication	**2**	42.961	2.294	0.881*	0.372	1782.765	214.255	14.135**	0.36
Genotypes	**16**	668.669**	1373.539**	1.344**	4.736**	11616.657**	1662.002*	13.884**	4.126*
Error	**32**	84.419	170.94	0.216	0.398	1829.536	591.172	2.783	1.276
h^2 in broad sense		69.76061	70.10526	63.51351	78.41649	64.06975	37.64767	57.07455	42.67745

* Significant at 5 % **highly significant at 1 %

in broad sense were showed especially under Sudr conditions. But under Inshas conditions the large environmental error was obvious therefore, the selection under Sudr conditions were fine and sensitivity. These results confirmed that, the large variations between induced mutant lines were occurred and they permit to selection of promising mutant lines under the two different adverse conditions.

Average mean of mutant lines and their parents for number of pods /plant and seed yield/plant under different conditions at M4 and M5 generations formed (Table 3). These results confirmed the above conclusion about importance of selection under Ras-Sudr conditions by comparison of Inshas conditions. Therefore, promising mutant lines were detected under Ras-Sudr conditions possessed significant at both M4and M5 generations, i.e., lines 8,18,38.63. Under Inshas conditions only line 75 alone had significant at M4 and M5 generations for number of pods/plant and seed yield/plant. Reflected behavior were recorded of some mutant lines at M4 and M5 generations, especially for number of pods/plant at M5 generation lines 5, 11, 22, 66, 87 under Ras-Sudr conditions. Line 66 alone possessed good performance under both conditions. These results confirmed that the genetic control of adverse environmental conditions was different between them and probable polygene mechanisms play an important role in the inheritance of saline and harsh conditions.

Oil Characters and Oil Percent

Mustard oil has characteristic pungency due to presence of glucosinolates in the seed meal and high erucic acid in oil, which renders mustard oil as less preferred over other cooking oils. None of the released varieties in India have the desired quality characters. Erucic acid content in the Indian cultivars is high averaging 49% compared to 25% in the European cultivars. The amount of glucosinolates varies from 150-240 μ mole/g of defatted meal. (Khalatkar and Indurkar, 1991) had indicated high effectiveness of gamma radiations and EMS in the induction of mutations in genes controlling the synthesis of erucic acid and glucosinolates. Besides classical breeding, mutation breeding has demonstrated the plasticity of seed oil quality with significant alteration in fatty acid composition and no apparent detrimental effects on the crop agronomics (Bhatia, 1991 and 1999).

In this study oil percent mutant lines seeds under both conditions was evaluated (Table 4), two lines 10, and 22 possessed significant at M4 and M5 generations under both conditions, while lines 8 and 75 had significant oil percent under Ras-Sudr conditions at M4 and M5 generations, as well as lines 5,66 and 74 had significant value of oil percent at M4 and M5 generations under Inshas conditions.

Acid value and flowering date were recorded at Table (5). These results confirmed that the acid value small than the unity, it was about between 0.4-0.633 and subsequently good oil were recorded for human nutrient. Flowering date confirmed that very importance of lines 11.38,63,66,74,75, whereas, it considered earliness criteria.

The conclusion of these results confirmed the importance of line 11 and 66 under both conditions. Line 38 and 63 considered as very important genotype of canola under Sudr conditions. While, The line 74 and 75 are very important genotypes under Inshas conditions. The performance of these lines was shown at Fig. 3.

These promising mutant lines require at next study to evaluation of them under large scale and detection of molecular markers as finger printing. As well as, more studies on oil criteria.

Table 3. Average mean least significant deference (LSD) for number of pods /plant and seed yield g/plant at Ras-Sudr and Inshas locations

	(Ras-Sudr)		(Inshas)		(Ras-Sudr)		(Inshas)	
	Number of pods		Number of pods		Seed yield g/ plant		Seed yield g/plant	
Line	M4	M5	M4	M5	M4	M5	M4	M5
Serow4	63.3	79	84.7	75.7	3.77	5.03	4.25	3.25
5	84.7*	94.7	148.3	85	4.60*	7.29*	6.08	4.96
8	83.3*	114.7*	183.0*	80	5.67*	8.50*	7.37*	4.92
10	76.7	88	145.3	112.3	4.99*	6.45*	6.77	6.69*
11	100.0*	91.7	191.7*	104.3	5.40*	6.99*	8.52*	6.56*
18	85.0*	106.0*	117	82.7	5.53*	6.87*	4.49	4.75
22	107.0*	84.7	83.7	82.7	5.06*	6.26*	5.55	4.93
X¯	85.7	94.1	136.2	89	5	6.77	6.15	5.15
Serow6	65.3	64.7	46	46.7	3.87	4.49	2.4	2.53
38	80.7*	92.3*	66	63.7	5.13*	6.50*	3.68	4.43*
X¯	73	78.5	56	55.2	4.5	5.5	3.04	3.48
Bactol	60.7	79.7	37	62.7	4.23	4.89	2.22	3.8
63	98.3*	151.0*	76	81.7	5.50*	7.91*	4.89	5.59
66	82.0*	78	139.3*	133.0*	5.43*	5.68	6.17*	5.52
X¯	80.3	102.9	84.1	92.5	5.05	6.16	4.43	4.97
Evita	62.3	71	74	65.7	3.4	4.43	2.51	3.73
74	65	90.3	148.7	97	4.70*	5.78	6.73*	5.88*
75	65	84.7	279.7*	107.0*	5.00*	5.24	10.07*	5.80*
87	70	121.0*	176.3*	114.0*	5.10*	7.60*	5.92*	8.89
92	71.7	70.7	124.3	121.7*	5.67*	4.7	6.54	6.51*
X¯	66.8	87.5	160.6	101.1	4.77	5.55	6.35	6.16
LSD 0.05	15.3	21.7	71.1	40.4	0.77	1.09	2.78	1.88

* Significant at 5% **highly significant at 1 %

Table 4. Average mean of seed oil percent of mutant lines compared to their parents in the two successive generations grown under the two locations

Line	Oil % (Sudr)		Oil% (Inshas)	
	M4	M5	M4	M5
con.1	43.24	38.7	39.43	42.73
5	43.85	38.78	42.17*	48.52*
8	45.71*	43.14*	40.62	48.58*
10	46.35*	46.30*	43.22*	46.69*
11	47.66*	41.72	40.76	46.76*
18	44.42	40.4	40.16	45.64
22	47.43*	43.63*	44.52*	46.67*
X¯	45.52	41.81	41.55	46.51
Con.2	44.46	42.57	39.83	42.52
38	46.25	44.67	40.78	48.20*
X¯	45.36	43.62	40.31	45.36
Con.3	39.93	39.59	39.18	45.4
63	44.85*	42.19	41.41	48.62*
66	46.67*	43.18	42.23*	48.33*
X¯	43.82	41.65	40.94	47.45
Con.4	41.74	41.70	41.48	43.85
74	44.20*	44.48	45.32*	49.79*
75	45.73*	46.17*	42.78	47.58*
87	42.64	44.65	43.45	47.15
92	42.52	41.47	43.76	46.66
X¯	43.37	43.69	43.36	47.01
LSD 0.05	2.1	3.80	2.3	3.60

* Significant at 5% **highly significant at 1%

Table 5. Average mean for acid value and flowering date at M5 generation of mutant lines

	Acid value		Flowering date / day	
line	**Inshas**	**Sudr**	**R1**	**R2**
Serow 4	0.64	0.519	70	70
line 5	0.4	0.381	85	80
Line 8	0.436	0.585	85	85
line10	0.622	0.503	80	80
line11	0.4	0.661	65	65
line18	0.579	0.514	87	85
ine22	0.44	0.473	85	85
X¯	0.502	0.519		
Serow6	0.629	0.597	80	80
line38	0.657	0.612	75	75
X¯	0.643	0.605		
Bactol	0.514	0.494	80	80
line63	0.588	0.6	60	60
line66	0.48	0.629	70	70
X¯	0.527	0.574		
Evita	0.428	0.509	85	85
line 74	0.544	0.547	65	65
line75	0.622	0.633	60	60
line 78	0.511	0.622	87	87
line92	0.514	0.518	80	80
X¯	0.524	0.566		

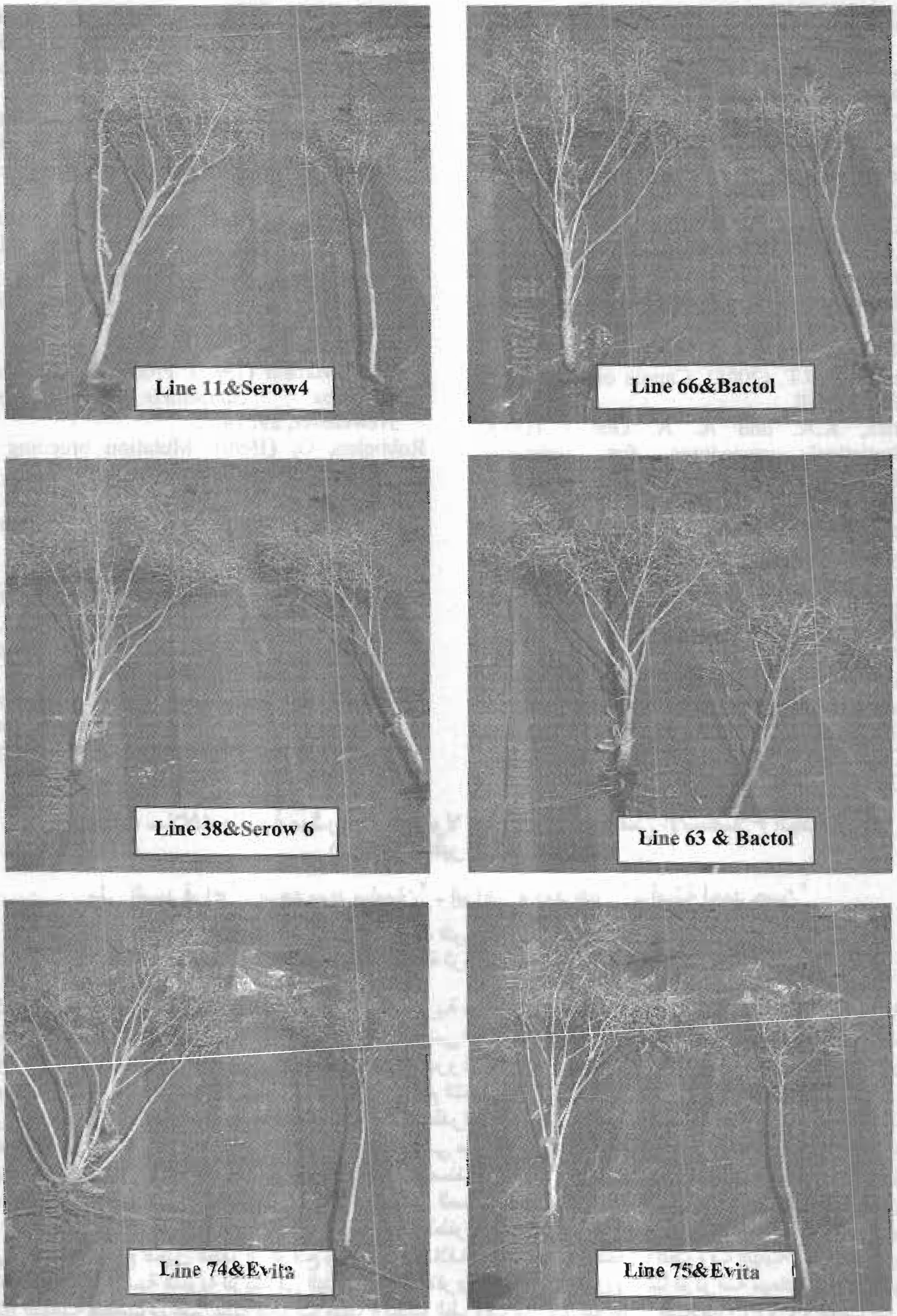

Fig. 3. The performance of 6 promising mutant lines and their control grown under: both conditions (lines 11 and 66), Ras- Sudr(lines 38 and 63) and Inshas (lines 74 and75)

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استحداث سلالات طفرية مبشرة في الكانولا تحت ظروف أراضى الإستصلاح الجديدة (الرملية الفقيرة، الملحية)

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كان الهدف من هذه الدراسة هو استحداث سلالات طفرية مبشرة فى الكانولا(محصول زيت) تمتلك محصول عالى البذور و محتوى عالى من الزيت و ذلك تحت ظروف أراضى الاستصلاح الجديدة فى رأس سدر- سيناء (ملحية) وإنشاص (رملية فقيرة). تم معاملة بذور الكانولا لأربع أصناف هى سرو٤ وسرو٦ و باكتول(محلية) وإفيتا (صنف مستورد) بأربعة جرعات من أشعة جاما هى: (0,100, 400, 600 Gy). تم انتخاب ٣٠ طفرة فى الجيل الطفرى الثانى على أساس عدد القرون لكل نبات و كذلك التغيرات المورفولوجية. أكدت طفرات الجيل الطفرى الثالث أن السلالات الطفرية المستحدثة تمتلك عدد كبير من القرون ومحصول البذور لكل نبات أعلى من الأم، وهذه السلالات الطفرية هى: ٨ ، ١١ ، ١٨ ، ٢٢ للصنف سرو٤ ، ٣٨ و ٤٥ للصنف سرو٦ ، ٦٣ و٦٦ للصنف باكتول ، ٧٤ للصنف إفيتا. وكان أعلى عدد قرون لكل نبات هو ١١٠ والذى سجل للسلالة الطفرية ٧٤ الناتجة من الصنف افيتا. كانت نتائج محصول البذور لكل نبات تسير فى نفس اتجاه عدد القرون لكل نبات، حيث وجد أن السلالات الطفرية التى تمتلك عدد قرون كبير تمتلك أيضا محصول بذور عالى. أظهرت نتائج الجيل الطفرى الرابع والخامس اختلافات كبيرة فى استجابتها للظروف البيئية المختلفة (موضع الدراسة). تم تقييم النسبة المئوية للزيت فى البذور للجيل الطفرى الرابع و الخامس وأيضا تم دراسة ميعاد التزهير. أظهرت النتائج النهائية الحصول علي ستة طفرات مبشرة تحت الظروف موضع الدراسة، السلالتين ١١، ٦٦ تحت ظروف المنطقتين (راس سدر وإنشاص) والسلالتين ٣٨ ، ٦٣ تحت ظروف راس سدر والسلالتين ٧٤ ، ٧٥ تحت ظروف انشاص .