

Studies on Genetic Selection to Produce New Genotypes of Mung bean (*Vigna Radiata* L.) by Gamma Rays and SCoT Marker

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ABSTRACT

A power of genetic selection is for producing new pure strains of interest species. The interest genes that are specifically expressed must be identified to develop the criterion of selection. The short period of Mung bean development could adapt to a broad sense of different factors such as agro- and eco-systems, plant rotation, plantation, and variety of germplasm. Three field experiments were carried out under the condition of Egyptian soil between Branch of Genetics, Department of Agricultural Plant, Faculty of Agriculture, Cairo, Al-Azhar University and Genetic Origins Department Research, Field Crops Research Institute, Agricultural Research Center, Bahtem Station, Qalyubia, Egypt on Mung bean. The genetic improvement and cultivars production of four strains of Mung bean was studied using gamma rays through selection and molecular studies techniques. The interaction between Mung bean strains and exposure to a dose of 40Gy of gamma rays had a significant effect on the characteristics of plant height (cm), number of branches/plant, number of pods/plant, number of seeds/pod, Pod length (cm), number of seeds/plant and total protein (%) at the level of significance ($P < 0.05$). The present research can provide the information of morphological and molecular characters of Mung bean. It is hoped that these findings are useful in new Genotypes confirmation. The position of bands by SCoT will provide the characters of identification of this plant.

Keywords: Genetic Selection, Mung bean, Gamma Rays, SCoT marker.

INTRODUCTION

Mung bean (*Vigna radiata* L.; Fabaceae), known green gram, is most important of the genus *Vigna* that is divided into seven subgenera owing over 150 species. Mung bean is very important leguminous crops in Egypt. Mung bean is an important grain legume because it is a rich source of proteins, vitamins and minerals (Smith, 2013). Cytological studies have indicated that basic chromosomal number ($2n = 22$) is same in almost all the varieties and species. The previous morphological description proved that the Mung bean germplasm has a low level of genetic diversity in respect of a set of gene pools (Ramanujam *et al.*, 2014). The short period of Mung bean development could adapt to a wide of various conditions like agro- and eco-systems, plant rotation, diversity of genotypes, and plantation manures. The morphological description is of utmost importance of characterization, classification, selection for desired germplasm, and collection for breeding program and genetic improvement (Kaga *et al.*, 2012). Agriculture is increasingly being studied extensively to produce more food, especially on the limited land resources of the 9 billion people expected on the planet by 2050.

The stats have been a gene-bank depository of 43,000 Mung bean germplasm, 110 varieties released by the World Vegetable Center (Southeast Asia), encompassing 415 cultivated (*Vigna radiata* var. *radiata*), 189 wilds (*Vigna radiata* var. *sublobata*), and 11 intermediate accessions from diverse geographic regions have been characterized using 19 azuki bean SSRs (Sehrawat *et al.*, 2015). The utilization of Mung bean gene-pools must be used with those germplasms by taking advantages of genetic diversity. Genetic improvement depends on crossing of elite parents with maximum genetic marker. It is necessary to state the nature and extent of the genetic diversity (Sanghani *et al.*, 2015).

The codon-targeting molecular marker (SCoT) is a useful marker for assessing the genetic variation (Collard & Mackill 2008) due to the fact there is no need for prior sequences information and high polymorphism and reproducibility. The SCoT marker depends on the conserved region surrounding the translation initiation codon, ATG (Sawant *et al.*, 1999). Due to lesser recombination intensity between SCoT marker and the gene of interest, it could directly use in marker-assisted breeding programs unlike random markers (Satya *et al.*, 2015). The SCoT has been successfully assessed for genetic variance, biodiversity, identification,

quantitative trait locus (QTL) mapping, and DNA fingerprinting. It is applied in various species such as date palm (Al-Qurainy *et al.*, 2015), Pistacia species (Sorkheh *et al.*, 2016), mango (Kuhn, 1978), and jojoba (El-Hak, 2019). The possibility of obtaining new cultivars through selection at a dose of 40Gy for the four lines compared with the parent strains (0.0 Gray) and the V.2010 variety by studying the characteristics for yield, seed protein content and the molecular level with SCoT markers.

MATERIALS AND METHODS

The present investigation was conducted during the three seasons 2017, 2018 and 2019. The three field experiments were carried out under the condition of Egyptian soil cooperating between Genetics Division, Agricultural Plant Department, Faculty of Agriculture, Cairo, Al-Azhar University, Department of Genetic Origins Research, Field Crops Research Institute (FCRI), Agricultural Research Center (ARC), Bahteem Station, Qalyubia, Egypt, on (*Vigna radiata*). The genetic improvement and cultivar production of four strains of Mung bean (*Vigna radiata*) were studied using gamma rays through selection and molecular studies techniques.

Yield characterization: Irradiation of seeds:

Seed irradiation of four strains of Mung bean is under study that had been exposed to three different doses (Cs137) of gamma irradiation (20, 40 & 60Gy) in addition to 0.0Gy as a control treatment (Abd El-Rahman *et al.*, 2016). The best treatments were 40 for the four strains. The best treatments were multiplied in the Genetic Resources Research Department, Field Crops Research Institute (FCRI), Agricultural Research Center (ARC), Bahteem Station, Qalyubia, Egypt during the 2017-2018 and 2109 growing seasons.

Mung bean lines and varieties:

Four Mung bean lines *Vigna radiata* (L.1, L.4, L.5, & L.21) differ in morphological properties and origin were studied and evaluated under Egyptian conditions as well as control local variety V.2010 (table 1).

Planting method:

All strains of Mung bean and the local cultivar were planted according to the recommended seeding rate as follow; 20 kg/acre for the L.1 and L.5 lines, 25 kg/acre for the line L.4 and V.2010 variety and family line L.21 was planted at a rate of 30 kg / feddan on two ploughs. Also, 2-3 seeds per hill were manual (dry seed in dry soil then watering)

with 20 cm interval on May 20, 31 and 21 in 2017, 2018, and 2019 seasons respectively. The harvest was on August 31 and 30 in the 2017 and 2018 seasons, respectively. While it was September the 2nd in the 2019 season.

Yield and yield components of Mung bean plant:

At the time of harvest, the following measurements were recorded as six selected random plant samples that were drawn from each plot to determine the following traits: Plant height (cm), number of branches per plant, number of pods/plant, number of seeds per pod, pod length (cm) and number of seeds per plant,

Quality characterization:

A calculable approximation for the standards of biochemical arrangement for the harvested grains from each sample of the studied Mung bean lines remained attained on wavelength area as of ~750nm to 2500nm of the electromagnetic range, by means of Near-Infrared (NIR) Spectroscopic analysis device, perfect DA1650, which mass-produced through FOSS Establishment. The estimate of the Biochemical Arrangement remained attained by the vital test site, faculty of agriculture, Al-Azhar University.

Statistical analysis:

Statistical analysis was performed using Statistical Package of Social Science, SPSS V.25 and computer program Microsoft Office Excel, 2019). The results were expressed as mean \pm standard deviation. The data were subjected to analysis of variance, ANOVA F test. Differences were considered significant at $p \leq 0.05$.

Molecular studies:

DNA Extraction:

Total genomic DNA of all the Mung bean selected seeds was obtained by DNeasy Mini Kit (QIAGEN). The concentration of DNA

was then determined based on a comparison of the DNA

samples with standard lambda DNA on 1% (w/v) agarose gel, after which it was adjusted to 5ng/ μ l.

Start Codon Targeted Polymorphism (SCoT):

The SCoT readers remained selected as of Collard & Mackill (2009) designed primers (table 2). These primers were synthesized by Operon Biotechnologies, Inc., GmbH, Cologne, Germany.

SCoT (Start Codon Targeted) Technique:

Five SCoT primers were used in this study (Table 2). The reaction conditions were optimized and the following reagents were mixed in a final volume of 25 μ l containing 2 μ L of template DNA (25 ng/ μ L); 2.5 μ L primer at 10 pM; 0.5 M dNTPs at 10mM; 0.15 μ L Taq DNA polymerase at 5 U/ μ L; 5 μ L 5X PCR buffer; and 14.75 μ L double distilled water. SCoT-PCR amplification was carried out in a T100- Bio-Rad Gradient Thermal cycler programmed as follows: initial denaturation was carried out at 94° C for 5 min, followed by 35 cycles of 94° C for 1 min, 50° C for 1 min, 72° C for 1.5 min, and final extension at 72° C for 7 min. The amplification products were separated in 1.5% agarose gels containing 0.5 μ g/mL of ethidium bromide through electrophoresis in 1X TBE buffer solution at 5 V/cm and photographed by Molecular ImagerR Gel Doc™ System with Image Lab™ Software, Bio-Rad.

RESULTS AND DISCUSSION

The interaction between strains of Mung bean and the dose of 40 gray of gamma rays:

The interaction between Plant height (cm) and number of branches/Plant of Mung bean and the dose of 40 gray of gamma rays:

The ANOVA analysis indicated that the interaction between Mung bean strains and exposure to a dose of 40Gy of gamma rays had a significant effect on the characteristics of plant height (cm) at the equal of implication ($P < 0.05$). The results showed that the highest values for plant height trait (106, 108 and 102.5 cm) were achieved by sowing seeds of strain L.1, which were treated at the dose of 40Gy in the three study seasons, respectively. While the lowest values for this trait (57 & 59 cm) were recorded in the cultivation of both strain L.1 and L.4 control during the 2017 and 2018 study seasons, while it was (77 cm) in the 2019 season for strain L.21, which were treated at the dose of 40Gy in compared to other transactions. The results also showed that the highest values for number of branches/plant trait (4.6, 5 and 6.5) were obtained by planting the seeds of the strain L.5 control during the three growing seasons, respectively. On the contrary, when planting the seeds of both control strain L.1 and L.21, as well as L.21, treated with the dose of 40Gy, (Table 3).

In the same line, Abd El-Rahman *et al.*, (2016) indicated that interaction effect between Mung bean lines and gamma doses radiation significantly affected Mung bean plant height

and number of branches per plant. The direction of the results shows that growing seeds of Fam.1 line that radiated with D40Gy and D60Gy gave the significant greatest values of plant height (85 and 124 cm) in 2013 and 2014 seasons, respectively as compared with the other treatments. On the other side, the shortest plant height was recorded by growing seeds of Fam.4 line which radiated with D40Gy (41.97cm) and Fam.21 line that is not radiated (D00) by gamma rays (75.50 cm), in 2013 and 2014 seasons, respectively. the growing seeds of Fam.1 line that radiated with D40Gy and non-irradiated (D00Gy) by doses of gamma rays scored the greatest values for number of branches per plant (2.67 & 4.33) in 2013 and 2014 seasons, respectively, as compared with the other treatments. While Fam.4 line and Fam.5 line that irradiated with 20Gy gave the lowest number of branches for Mung bean plant (1.00 and 1.00) during the seasons of 2013 and 2014, respectively. This trend is in harmony with previous results reported by Tah *et al.*, (2006) who observed that two varieties of Mung bean (K851 and Sonja) were found to have a maximum petiole length in plants treated with a lower dose of gamma irradiation. Also, Rahman *et al.*, (2015) showed that radiation effects in relation to plant height showed significant differences. The value of plant height at 20 kR was found insignificant with the control which indicates that the plant height was not much influenced by 20 kR. The values of other treatments were significantly different from the controls but insignificant within the radiation levels. However, the results showed that with the increase in doses of gamma rays the plant height was reduced in all the genotypes. A linear negative trend due to application of gamma rays remained stated by many labors (Sarwar *et al.*, 2015; Shamsheerband *et al.*, 2020) in Mung bean and black gram. Additionally, El-Degwy & Hathout, (2014) found that the radiation doses of 5 and 10 Kr have slightly reduced plant height while other doses had no considerable effect on plant height.

The interaction between pods/plant and number of seeds/pod of Mung bean and the dose of 40 gray of gamma rays:

The results indicated a significant effect of the spontaneous interaction between Mung bean strains and the dose of 40Gy of gamma rays on the characteristic of the numbers of pods/plant in the precipitating seasons. Whereas the highest values were (32.6, 44.4 and 153.5), which were achieved by planting the seeds of both strains L.1 and L.4 treated at

the dose of 40Gy, as well as the control basket L.1 in the three seasons, respectively. While the control L.5, L.4 and L.5 strains with a dose of 40Gy achieved the lowest values in the three seasons, which are (7, 12.6 and 47.5), respectively. As for the number of seeds/pod, the cultivation of the seeds of the strain L.21, which was treated with radiation dose 40Gy led to the highest values (12.4 and 11.8) for this trait in both seasons 2017 and 2018, while in the 2019 season the control L.4 strain had the highest values compared to the other strains in the study, which were (13), which were less of the V.2010 class in that season, where it scored (13.6). Also, planting seeds of control strain L.21 gave the lowest values for this trait (9.4 and 9.6) during the 2017 and 2018 seasons, respectively. While the lowest values were in the 2019 season when planting seeds of strain L.1 with a dose of 40Gy, which was recorded (11). (Table 4).

These results were in agreement with Abd El-Rahman *et al.*, (2016) who concluded that growing seeds of Fam.1 line that radiated with D20 Gy and D40 Gy gave the significant greatest values for number of pods/plot (1357.00 and 2634.67) in both seasons. On the other hand, growing seeds of Fam.5 line that irradiated with D40 Gy in 2013 and 2014 seasons scored the lowest values for that trait (328.67 and 650.00) and the differences between them reached to the significant level (5%). Similar results confirmed by Ameen *et al.*, (2006) who concluded that dry seeds of four pea varieties were treated with gamma rays (0.0, 5, 10 and 15 Kr) on the dose of 5 Kr increased the mean square values for number and weight of pods per plant. On the other hand, the increasing of dose level gave the lowest mean square values for most studied characters. Khan *et al.*, (2019) screened the high-yielded mutants of *Vigna radiata* induced by lower doses of biochemical mutagens in number of pods/plant and total seed harvest. Ali *et al.*, (2015) reported that gamma rays affect leaf canopy and seed harvest, particularly those owing large leaf in size that expose to photosynthesis resulting in high yield frequency.

The interaction between Pod length (cm), numbers of seeds/plant and total protein (%) of Mung bean strains and the dose of 40 gray of gamma rays:

The obtained results indicated that by planting the seeds of the strain L.21, which were treated with the radiation dose of 40Gy, the highest values of the Pod length compared to each of the other studied strains in both

seasons 2017 and 2018, where it recorded (12 and 11.6 cm) on respectively, while the L.4 strain with a dose of 40Gy had the highest values for that trait in the 2019 season (10.4 cm), which was lower than the V.2010 variety recorded in that season (12.5 cm). As for the number of seeds/plant, the highest values were recorded by the strains L.1 and L.21 treated at the dose 40Gy, as well as the control L.1, where the values were (348, 323, and 1773.7) for the three seasons, respectively, compared to the rest of the strains and the cultivated variety. As for the total protein (%), the highest values for the protein content of seeds were (29.05%, 29.08 and 30.12%) which were recorded by the strain L.21 with a dose of 40Gy in the three seasons, respectively. While the lowest values were (25.64%) recorded by the control L.1 strain in the three seasons, respectively. (Table 5).

Our results disagree with Astuti *et al.*, (2000) who reported that protein content, essential amino acids, minerals, trace elements, and vitamins do not be lost significantly through an over dose of 10 kGy. It was found that the protein contents at a dose of 608.80 Gy (23,23±0,12) was higher than at a dose of 222.20 Gy (21,90±0,22%) and at zero dose of control (22,16±0,26%). Jan *et al.*, (2012) reported that lysine and histidine of *Ginseng panax* were unaffected the irradiation by gamma at a dose of 5 kGy. Al-Jassir, (1992) investigated that the content of arginine, methionine, lysine, phenylalanine, and leucine of *Allium sativum* L. increased slightly; while others reduced especially under a high doses.

Molecular Study:

Amplification of SCOT-PCR parameters and phenotypic polymorphism on genotypes

The results indicated that the total average of the amplified fragments of the initiator SCoT -1 primer was (13 bands), the molecular weight was between (160 - 823 base pairs) and the total average of the number of bands was (51 bands), while the number of the most visible distinct bands was (5 bands). It was characterized by (strain L.1 when exposed to radiation dose 40Gy at molecular weight 637 and 160) and (strain L.4 control at molecular weight 181) as well as (strain L.21 control at molecular weight 188 and 163). The polymorphism rate was 84,615%. SCoT 2-primer was (23 bundles), the molecular weight was between (271-1260 base pairs) and the total number of bundles was (75 bundles), while the most visible distinct bundles were (13 bundles characterized by (strain L.1 control

at molecular weight 401-457-532-719) and (L.4 strain at the dose 40Gy at the molecular weight 1192) and (the control L.5 strain at the molecular weight 657) and (the L.5 strain treated with dose 40Gy at the molecular weight 521) and (Strain L.21 dosed 40Gy at the molecular weights 445-597-649) as well as (Class V 2010. at the molecular weights 427-569-1114). The percentage of polymorphism was 84,615%. The results showed that the number of amplified fragments of SCoT 3-primer was (8 bands) and the molecular weight was between (186-617 base pairs), while the average number of total bands was (39 bands), and no distinct bands were observed in all strains in all treatments. The polymorphism rate was 75,000%. SCOT 4-primer was (17 bands) and the molecular weight was between (327 - 1440 base pairs), while the average number of the total bands was (48 bands). It was characterized by (strain L.1 control at molecular weights 399-520-696-773-892-1440) as well as (strain L.1 with dose 40Gy at molecular weight 1164). The polymorphism rate was 96.118%. The results indicated that the average number of amplified fragments of the SCOT-5 primer was (19 bundles), and the molecular weight was between (184-1096 base pairs). The average number of total bands was (42 bundles), while the number of distinct bands most visible was (12 bands), which were characterized by both (L.1 control strain at molecular weight 276), (strain L.1 with a dose of 40Gy at molecular weights 296-360), (strain L.4 control at molecular weights 545-995), (strain L.4 dose-treated 40Gy at molecular weight 348), (strain L.5 control at molecular weights 508-605-696) and (strain L.5 dose 40Gy at weights at molecular weights 331-417) as well as (L.21 control strain at molecular weight 687). It was a multiplicity (Tables 6, 7 and figures 1, 2, 3, 4 and 5).

In a study by Mahdy *et al.*, (2021) SCoT polymorphism for polymorphic screening seven primers of SCoT were used. The analysis of genotypes with seven SCoT primers provided a total of 66 amplicons with a range between 178.2 to 1351bp. There were 66 bands, ranging from 6 (Primer SCoT-2 and SCoT-11) to 14. (Primer SCoT-10). In addition, the seven primary molecular markers are classified as positive molecular markers (41 molecular markers), which are compared with controls of several molecular dimensions in irradiated callus. On the contrary, two negative molecular markers with molecular sizes 940 and 400 bp were found only in control in comparison with irradiated callus. In

addition, in both control and irradiated callus 25 common molecular markers were identified. It is predicted that the SCoT markers are connected to functional genes and corresponding characteristics, so that the amplicons can be translated to gene target marker systems. Also, these markers are multilocuses that are useful in achieving high genetic polymorphism. These changes with gamma will correlate with the photoproduct levels of the Taq polymerase binding sites after radiation in the DNA template. Furthermore, the results obtained suggested that the SCoT marker can be used to effectively evaluate the variation between treated *Atropa belladonna* (Xiong *et al.*, (2013), EL-Shaer & Ibrahim (2021); Vanmathi *et al.*, (2021) analyzed the genetic variation among M1, M2, and M3 through the SCoT marker. Seven primers gave high reproducible comparing to primer SCoT-09 that gives no amplification. About 133 bands out of 222 bands were polymorphic which ranged from 3.03–96.07%, with a mean of 67.10%. a total number of polymorphic amplicons varies from 2 band for SCoT-01 to 49 bands for SCoT-06, with an average of 19%. The SCoT-01 scored the highest primer, while the smallest number of amplicons screened in SCoT-10. The major polymorphic bands were 49 bands for SCoT-06 and the minor ones were 2 bands for SCoT-01. The SCoT-03 and SCOT-08 primers scored absent of bands for M3 comparing to M1 and M2 generations. In M2 generation, all the primers of M2 gave a full of amplicons, while SCOT-08 primer did not give any amplicons for M3 generation. It may be due to a problem occurring during PCR reaction, DNA degradation by irradiation, or mismatches.

Polymorphism (%) among five random primers using SCoT-PCR:

The results indicated that (83 bundles) were produced. The average percentage of phenotypic polymorphism was recorded 86.617%, ranging from 75.00% to 94.73%. In addition to the presence of distinct bands representing 25.57% in the initiator SCoT-5, while the initiator SCoT-3 showed the absence of any distinct bands. While the polymorphism of the phenotype without distinct bundles was (84.6, 84.6, 75, 94.1, 94.7%) respectively for the previous prefixes, (Table 8).

Cluster analysis and similarity between 9 genotypes:

The results showed that the cluster analysis divides the nine genotypes of Mung bean into two groups at the distance (12.48). The first

cluster remained alienated hooked on 2 diverse clusters at the reserve (8.53). Below the first group contains one genetic structure (strain L.1 Control), Under the second group, which in turn was divided into two under-under groups at the distance (6.67). The first sub-group includes two genotypes (strain 1L. irradiated with a dose of 40Gy and strain L.4 control). The second group also includes two genotypes under the second group (strain 4L. Radiation treated with a dose of 40Gy and strain L.5 Control). As for another cluster, it remained alienated hooked on 2 diverse clusters at the reserve (6.67). Under the first group, it contains one genotype (variety 2010.V). Though another cluster remained alienated hooked on 2 clusters at the reserve (5.07). The first sub-group contains one genotype (strain 5L. Radiated at a dose of 40Gy). While the second group under - under the second group contained two genotypes (strain L.21 control as well as L.21 Radiation treated with a dose of 40Gy). (Figure 6).

CONCLUSION

The present study can provide the information of morphological characters of *Vigna radiata* (L.). It is hoped that these finding were useful in species confirmation. Genetic selection is still almost of promising methods for obtaining germplasm, especially selection among a large number of collections and gene pools. The target genes must be identified for genetic improvement. The short period of Mung bean development could adapt to a wide range of various conditions. The Start Codon target (SCoT) markers proved a promising technique for assessing genetic variation in plants due to matching start codon of genes.

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Table 1: Pedigrees of four strains of Mung bean (*Vigna radiata*) and a local type according to Genetic Resources Research Division, Field Crops Research Institute (FCRI), Agricultural Research Center (ARC), Bahteem Station, Qalyubia, Egypt.

No.	Geno type	Origin	Pedigree	Testa color	Hilum color	Testa shape	Pod color	Seed Size	100 seed /weight (g)
1	L.1	India	Line 63	Light Green	White	Smooth	Black	Small	2-3 g
2	L.4	India	Line 80	Light Green	White	Smooth	Black	Medium	4-5 g
3	L.5	India	Line 83	Yellow	White	Smooth	Black	Small	2-3 g
4	L.21	Egypt	V.C 2719	Dark Green	White	Rough	Black	Big	6-7 g
5	V.2010	V.2010 Egypt	Individual plant selection	Green	White	Smooth	Beige	Medium	4-5 g

Table (2): List of primer names and their nucleotide sequences which used for SCoT procedure.

No	Name	Sequence
1	SCoT-1	5'-CAACAATGGCTACCACCA-3'
2	SCoT -2	5'-CAACAATGGCTACCACCC-3'
3	SCoT -3	5'-CAACAATGGCTACCACCG-3'
4	SCoT -4	5'-CAACAATGGCTACCACCT-3'
5	SCoT -5	5'-CAACAATGGCTACCACGA-3'

Table 3: Effect of different lines and gamma irradiation as well as the interaction effect between them on plant height (cm) and number of branches/plants on Mung bean plant in 2017, 2018 and 2019 seasons.

Treatments		Plant height (cm)			Number of branches/Plant		
Lines	Radiation (Gray)	2017	2018	2019	2017	2018	2019
L.1	D.00	57 ^d	59 ^e	90 ^a	2 ^b	2 ^d	4.8 ^{bc}
	D.40	106 ^a	108 ^a	102.5 ^a	4.6 ^a	4.4 ^b	5.5 ^{abc}
	Mean	81.5 ^a	83.5 ^b	96.5 ^a	3.3 ^b	3.2 ^b	5.2 ^a
L.4	D.00	57 ^d	59 ^e	86.2 ^a	4.2 ^a	4.2 ^b	5.7 ^{abc}
	D.40	64 ^{cd}	74 ^c	82.5 ^a	4.6 ^a	4.6 ^{ab}	6.2 ^{ab}
	Mean	60.5 ^b	66.5 ^d	84.5 ^{ab}	4.4 ^a	4.4 ^a	6 ^a
L.5	D.00	64 ^{cd}	68 ^d	87.5 ^a	4.6 ^a	5 ^a	6.5 ^{bc}
	D.40	68 ^c	79 ^b	92.5 ^a	2.8 ^b	3.2 ^c	4.5 ^c
	Mean	66 ^b	73.5 ^c	90 ^{ab}	3.9 ^{ab}	4.1 ^a	5.5 ^a
L.21	D.00	71 ^c	70 ^d	87.5 ^a	2 ^b	2.2 ^d	5 ^{bc}
	D.40	90 ^b	106 ^a	77 ^a	2 ^b	2.6 ^d	4.2 ^c
	Mean	80.5 ^a	88 ^a	83 ^b	2 ^b	2.4 ^c	4.6 ^a
Mean	V.2010	75 ^a	75 ^a	95 ^a	2 ^c	2 ^c	4 ^c
Overall line x Radiation							
	D00	62.3 ^b	64 ^b	87.8 ^a	3.5 ^a	3.4 ^b	5.6 ^a
	D40	82 ^a	91.8 ^a	89 ^a	3.4 ^a	3.7 ^a	5.2 ^a

Table 4: Effect of different lines and gamma irradiation as well as the interaction effect between them on number of pods/plant and number of seeds/pods of Mung bean plants in 2017, 2018 and 2019 seasons.

Treatments		Number of pods/plants			Number of seeds/Pod		
Lines	Radiation (Gy)	2017	2018	2019	2017	2018	2019
L.1	D.00	11.8 ^d	14.8 ^e	153.5 ^a	10.2 ^{cd}	10.4 ^{bc}	12 ^a
	D.40	32.6 ^a	39.2 ^b	138 ^a	12 ^{ab}	10.4 ^{bc}	11.7 ^a
	Mean	22.2 ^a	27 ^a	145.8 ^a	11.1 ^a	10.4 ^a	11.9 ^b
L.4	D.00	10.6 ^{cd}	12.6 ^e	113.7 ^a	9.4 ^d	10.4 ^{bc}	13 ^a
	D.40	29.2 ^a	44.4 ^a	57 ^a	11 ^{bc}	11.6 ^a	12.7 ^a
	Mean	19.9 ^a	28.5 ^a	85.4 ^b	10.2 ^a	11 ^a	12.9 ^a
L.5	D.00	7 ^d	13.2 ^e	66 ^a	10.8 ^{bcd}	10 ^{bc}	12.2 ^a
	D.40	13.4 ^c	21 ^d	47.5 ^a	11.8 ^{ab}	10.8 ^{ab}	12.5 ^a
	Mean	10.2 ^b	17.1 ^c	56.8 ^b	11.3 ^a	10.4 ^a	12.4 ^b
L.21	D.00	8.2 ^d	14.4 ^e	64.5 ^a	9.4 ^d	9.6 ^c	12 ^a
	D.40	22.2 ^b	25.6 ^c	54.5 ^a	12.4 ^{ab}	11.8 ^a	12.2 ^a
	Mean	15.2 ^b	20 ^b	59.5 ^b	10.9 ^a	10.7 ^a	12.1 ^b
Mean	V.2010	12 ^a	21 ^a	65 ^c	9.4 ^b	11.8 ^a	13.6 ^a
Overall line x Radiation							
	D00	9.4 ^b	13.8 ^b	99.8 ^a	10 ^b	10.1 ^b	12.3 ^a
	D40	24.4 ^a	32.6 ^a	74.3 ^b	11.8 ^a	11.2 ^a	12.4 ^a

D*=dose, a, b, c & d = Statistical values

Table 5: Effect of gamma irradiation dose on different lines on pod length (cm), amount of seeds/plant and total protein (%) of Mung bean plants in 2017,2018 and 2019 seasons.

Treatments		Pod length(cm)			Number of seeds/Plant			% Protein		
Lines	Radiation (Gy)	2017	2018	2019	2017	2018	2019	2017	2018	2019
L.1	D.00	6.7 ^d	6.8 ^e	7.6 ^d	117 ^{cd}	204 ^b	1773.7 ^a	25.64	25.64	25.64
	D.40	8.4 ^c	9.6 ^{cd}	7.1 ^e	348 ^a	300 ^a	1475 ^a	26.48	27.81	28.78
	Mean	7.6 ^c	8.2 ^b	7.4 ^c	232.5 ^a	252 ^a	1624.4 ^a	26.06	26.06	26.73
L.4	D.00	10.2 ^b	10.6 ^b	10.2 ^{ab}	139.6 ^{cd}	110.6 ^d	1431 ^a	26.32	26.32	26.32
	D.40	9.8 ^b	9.4 ^{cd}	10.4 ^a	320 ^a	290 ^a	708.3 ^b	26.26	28.34	29.84
	Mean	10 ^b	10 ^a	10.3 ^a	229.8 ^a	200.3 ^b	1069.7 ^b	26.29	26.29	27.33
L.5	D.00	10 ^b	8.8 ^d	9.3 ^c	70 ^d	122 ^d	740.2 ^b	28.05	28.05	28.05
	D.40	11.7 ^a	11.6 ^a	10.1 ^{ab}	164 ^c	221 ^b	544.7 ^b	28.21	27.81	29.59
	Mean	10.9 ^a	10.2 ^a	9.7 ^b	117 ^b	171.5 ^b	642.5 ^c	27.93	28.13	27.93
L.21	D.00	8.3 ^c	10 ^{bc}	9.8 ^b	76 ^d	144.6 ^c	725.5 ^b	27.18	27.18	27.18
	D.40	12 ^a	11.6 ^a	9.5 ^c	244.2 ^b	323 ^a	595 ^b	29.05	29.08	30.12
	Mean	10.2 ^b	10.8 ^a	9.7 ^b	160.1 ^{ab}	233.8 ^a	660.3 ^c	28.13	28.12	28.13
Mean	V.2010	9 ^b	11.8 ^a	12.5 ^a	114 ^a	221.2 ^a	747.5 ^b	28.76	28.76	28.76
Overall line x Radiation										
	D00	8.8 ^b	9.1 ^b	9.3 ^a	100.7 ^b	150.3 ^b	1196.1 ^a	26.8	26.8	26.8
	D40	10.5 ^a	10.6 ^a	9.3 ^a	269.1 ^a	287 ^a	831.2 ^b	27.5	28.26	29.58

D*= dose, a, b, c & d = Statistical values

Table 6: Distribution and size of mono-morphic and polymorphic bands that obtained by SCoT analysis on *Vigna radiata* lines and local variety using primer SCoT-1, SCoT-2 and SCoT-3.

SCoT-1 Primer											
AF	MW	L. 1 line		L. 4 line		L. 5 line		L.21 line		V.2010	Polymorphism
		D00	D40	D00	D40	D00	D40	D00	D40		
		(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	
1	822.617	0	1	1	1	1	1	1	1	0	Polymorphic
2	637.931	0	1	0	0	0	0	0	0	0	Unique
3	455.21	1	1	1	1	1	1	1	1	1	Monomorphic
4	399.941	0	1	1	0	0	1	1	1	1	Polymorphic
5	354.646	0	1	1	1	1	1	1	1	1	Polymorphic
6	282.76	1	1	1	1	1	1	1	1	1	Monomorphic
7	238.305	1	1	1	0	0	0	0	0	0	Polymorphic
8	235.023	0	0	0	0	0	1	1	0	0	Polymorphic
9	188.252	0	0	0	0	0	0	1	0	0	Unique
10	182.258	1	1	0	0	0	0	0	0	0	Polymorphic
11	181.417	0	0	1	0	0	0	0	0	0	Unique
12	163.874	0	0	0	0	0	0	1	0	0	Unique
13	160.129	0	1	0	0	0	0	0	0	0	Unique
Total		4	9	7	4	4	6	8	5	4	51
SCoT-2 Primer											
1	1260.132	0	0	0	0	1	0	0	1	0	Polymorphic
2	1245.194	0	1	1	0	0	0	0	0	0	Polymorphic
3	1196.665	0	0	0	0	0	1	1	0	0	Polymorphic
4	1191.917	0	0	0	1	0	0	0	0	0	Unique
5	1114.030	0	0	0	0	0	0	0	0	1	Unique
6	719.431	1	0	0	0	0	0	0	0	0	Unique
7	656.570	0	0	0	0	1	0	0	0	0	Unique
8	648.786	0	0	0	0	0	0	0	1	0	Unique
9	621.027	0	1	1	1	0	0	0	0	0	Polymorphic
10	596.824	0	0	0	0	0	0	0	1	0	Unique
11	585.079	0	0	0	0	0	1	1	0	0	Polymorphic
12	569.022	0	0	0	0	0	0	0	0	1	Unique
13	531.838	1	0	0	0	0	0	0	0	0	Unique
14	521.372	0	0	0	0	0	1	0	0	0	Unique

15	457.273	1	0	0	0	0	0	0	0	0	Unique
16	444.724	0	0	0	0	0	0	0	1	0	Unique
17	442.959	0	1	1	0	1	0	0	0	0	Polymorphic
18	427.391	0	0	0	0	0	0	0	0	1	Unique
19	401.054	1	0	0	0	0	0	0	0	0	Unique
20	376.340	0	1	1	1	1	1	1	1	1	Polymorphic
21	334.031	1	1	1	1	1	1	1	1	1	Monomorphic
22	290.644	1	1	1	1	1	1	1	1	1	Monomorphic
23	271.652	1	1	1	1	1	1	1	1	1	Monomorphic
Total		8	9	8	8	8	8	8	9	9	75
SCoT -3 Primer											
1	617.464	0	0	0	1	1	1	1	1	0	Polymorphic
2	437.150	0	0	0	0	1	1	1	1	0	Polymorphic
3	348.853	0	1	1	1	1	0	0	0	0	Polymorphic
4	340.913	1	0	0	0	0	0	0	0	1	Polymorphic
5	331.623	0	0	0	0	0	1	1	1	0	Polymorphic
6	282.262	1	1	1	1	1	1	1	1	1	Monomorphic
7	218.106	1	1	1	1	1	1	1	1	1	Monomorphic
8	185.642	0	0	1	1	0	1	0	0	0	Polymorphic
Total		3	3	4	5	5	6	5	5	3	39

AF= Amplified Fragment, MW=Molecular Weight, 1=presence of band, 0= absence of band and V.2010= Cultivated variety

Table 7: Distribution, size of mono-morphic and polymorphic bands that obtained by SCOT analysis on *Vigna radiata* lines and local variety using primer SCoT-4 and SCoT-5.

SCoT - 4 Primer											
AF	MW	L. 1 line		L. 4 line		L. 5 line		L.21 line		V.2010 (9)	Polymorphism
		D00 (1)	D40 (2)	D00 (3)	D40 (4)	D00 (5)	D40 (6)	D00 (7)	D40 (8)		
1	1440.092	1	0	0	0	0	0	0	0	0	Unique
2	1302.928	0	0	1	1	1	0	0	0	0	Polymorphic
3	1163.538	0	1	0	0	0	0	0	0	0	Unique
4	1113.989	0	0	0	0	0	0	1	1	1	Polymorphic
5	892.260	1	0	0	0	0	0	0	0	0	Unique
6	772.897	1	0	0	0	0	0	0	0	0	Unique
7	756.261	0	1	1	0	0	0	0	0	0	Polymorphic
8	724.056	0	0	0	0	0	1	1	1	1	Polymorphic
9	696.245	1	0	0	0	0	0	0	0	0	Unique
10	613.695	0	1	1	1	1	0	0	0	0	Polymorphic
11	572.417	0	0	0	0	0	1	1	1	1	Polymorphic
12	520.155	1	0	0	0	0	0	0	0	0	Unique
13	476.796	0	1	1	1	1	0	0	0	0	Polymorphic
14	462.491	0	0	0	0	0	1	1	1	1	Polymorphic
15	359.322	1	1	1	1	1	1	1	1	1	Monomorphic
16	398.881	1	0	0	0	0	0	0	0	0	Unique
17	326.516	0	0	0	0	0	1	1	1	1	Polymorphic
Total		7	5	5	4	4	5	6	6	6	48
SCoT - 5 Primer											
1	1095.794	0	1	0	1	0	0	0	0	0	Polymorphic
2	1058.176	0	0	0	0	1	0	1	0	0	Polymorphic
3	995.425	0	0	1	0	0	0	0	0	0	Unique
4	900.309	0	0	1	0	1	1	0	0	0	Polymorphic
5	695.833	0	0	0	0	1	0	0	0	0	Unique
6	686.777	0	0	0	0	0	0	1	0	0	Unique
7	629.344	1	1	1	1	0	0	0	0	0	Polymorphic
8	610.398	0	0	0	0	0	1	1	1	1	Polymorphic
9	605.091	0	0	0	0	1	0	0	0	0	Unique
10	544.888	0	0	1	0	0	0	0	0	0	Unique

11	508.119	0	0	0	0	1	0	0	0	0	Unique
12	417.472	0	0	0	0	0	1	0	0	0	Unique
13	359.874	0	1	0	0	0	0	0	0	0	Unique
14	347.520	0	0	0	1	0	0	0	0	0	Unique
15	331.222	0	0	0	0	0	1	0	0	0	Unique
16	295.674	0	1	0	0	0	0	0	0	0	Unique
17	275.722	1	0	0	0	0	0	0	0	0	Unique
18	235.614	0	1	1	1	1	1	1	0	0	Polymorphic
19	183.699	1	1	1	1	1	1	1	1	1	Monomorphic
Total		3	6	6	5	7	6	5	2	2	42

AF= Amplified Fragment, MW=Molecular Weight, 1=presence of band, 0= absence of band and V.2010= Cultivated variety

Table 8: Polymorphism (%) among five random primers using SCoT-PCR

Bands	SCOT-1	SCOT-2	SCOT-3	SCOT-4	SCOT-5	Total	Mean
Molecular Weight (bp)	160.129 - 822.617	160.102 - 1260.132	185.642 - 617.464	326.516 - 1440.092	183.699 - 1095.794	-	-
Total (AF)	13	26	8	17	19	83	16.6
Monomorphic bands	2	4	2	1	1	10	2
Unique bands	5	13	0	7	12	37	7.4
Polymorphic (without Unique)	6	9	6	9	6	36	7.2
Polymorphic (with Unique)	11	22	6	16	18	73	14.6
Total number of bands	51	75	39	48	42	255	51
Polymorphism (%)	84.615%	84.615%	75.000%	94.118%	94.737%	75.00 - 94.73 %	86.617%

AF= Amplified Fragment

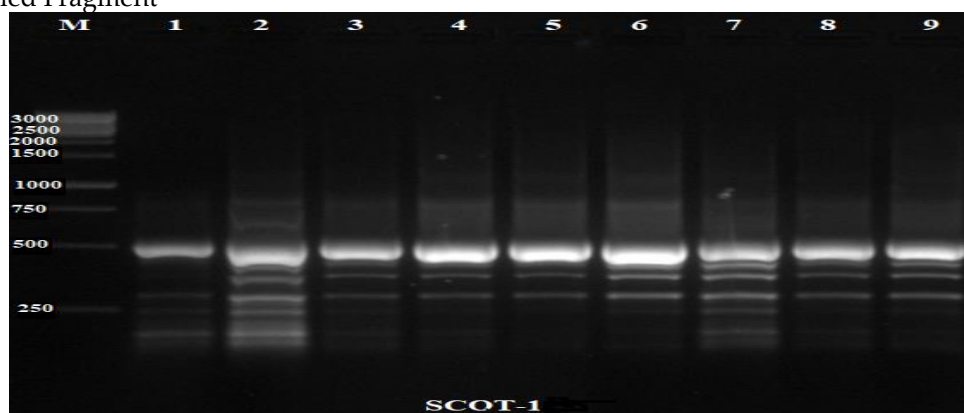


Figure 1: SCoT analysis profiles for the nine accessions of Mung bean plant generated by primer SCoT-1.

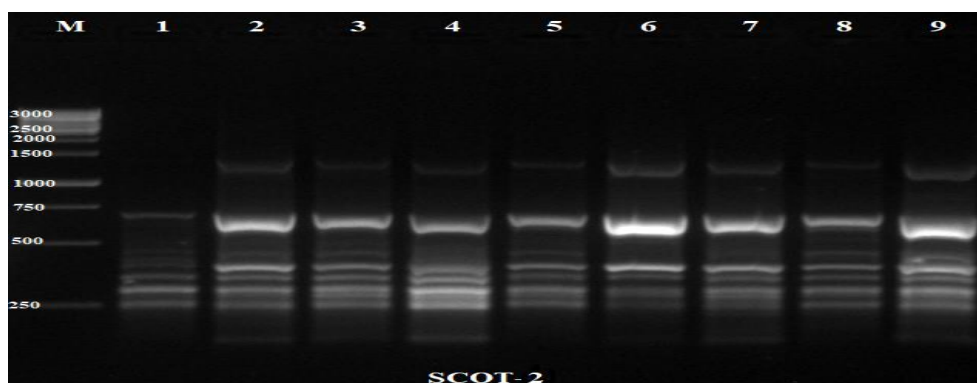


Figure 2: SCoT analysis profiles for the nine accessions of Mung bean plant generated by primer SCoT-2.

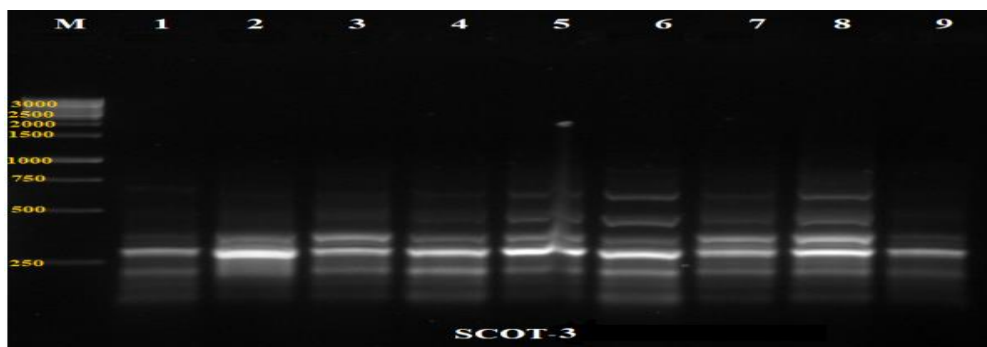


Figure 3: SCoT analysis profiles for the nine accessions of Mung bean plant generated by primer SCoT-3.

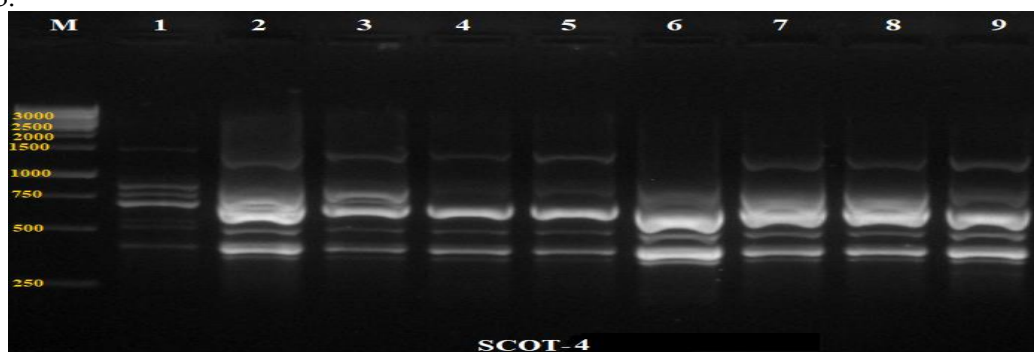


Figure 4: SCoT analysis profiles for the nine accessions of Mung bean plant generated by primer SCoT-4.

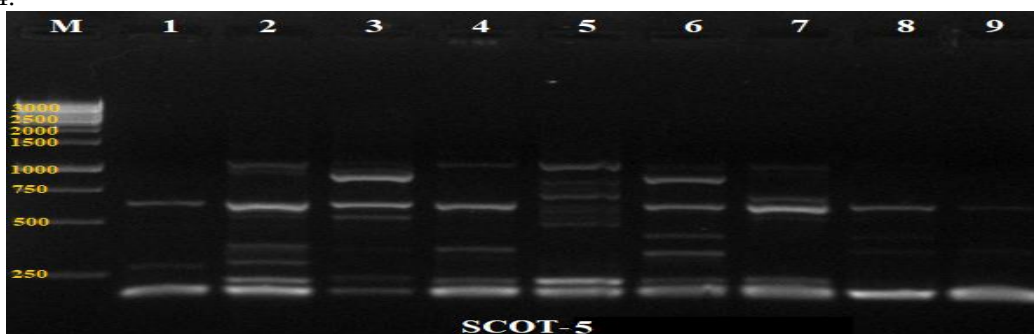


Figure 5: SCoT analysis profiles for the nine accessions of Mung bean plant generated by primer SCoT-5.

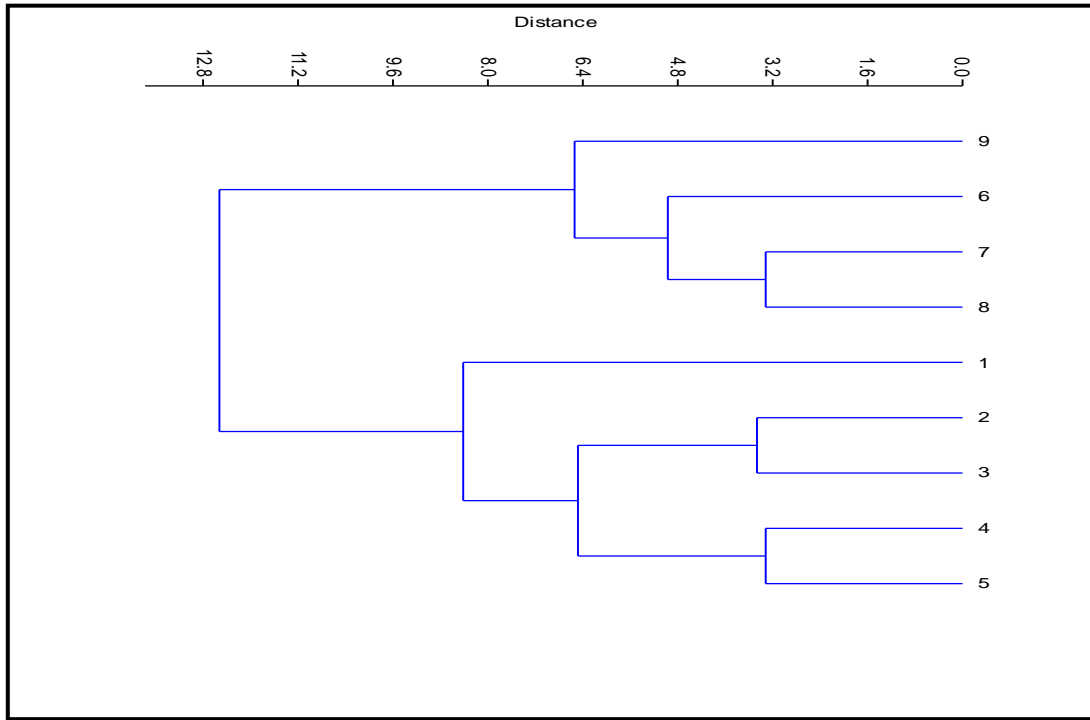


Figure 6: Dendrogram tree constructed using unweighted pair group method with arithmetic average (UPGMA, Jaccard similarity). Cluster analysis based on SCoT Markers; 1, 2= L.1 (D0, D40), 3,4 = L2 (D0, D40), 5,6=L.3(D0, D40), 7,8=L.4(D0, D40) and while 9 = V.2010.

دراسات على الانتخاب الوراثي لإنتاج تراكيب وراثية جديدة من فول المونج بواسطة أشعة جاما والواسم الجزيئي SCoT.

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الملخص العربي

يعد الانتخاب الوراثي أداة قوية للحصول على سلالات نقية أو شبه نقية من أنواع النباتات المتمايزة، لتطوير هذه الأداة يجب تحديد الجينات في نوع الخلية المطلوب والتي يتم التعبير عنها على وجه التحديد، تسمح فترة النمو القصيرة لفول المونج بالقدرة على التألف مع عدد كبير جداً من أنظمة المحاصيل والدورات الزراعية المختلفة وبالتالي التنوع في أنظمة الزراعة. أجريت ثلاث تجارب حقلية تحت ظروف التربة المصرية بين فرع الوراثة - قسم النبات الزراعى - كلية الزراعة بالقاهرة - جامعة الأزهر وقسم بحوث الأصول الوراثية - معهد المحاصيل الحقلية - مركز البحوث الزراعية - محطة بهتميم - القليوبية - مصر على فول المونج. تمت دراسة التحسين الوراثي وإنتاج أصناف من أربع سلالات من فول المونج باستخدام أشعة جاما من خلال الانتخاب وتقنيات الدراسات الجزيئية، كان التفاعل بين سلالات فول المونج والتعرض لجرعة 40 جراى من أشعة جاما له تأثير كبير على خصائص ارتفاع النبات (سم) وعدد الفروع/ نبات وعدد القرون/ نبات وعدد البذور/ قرن وطول القرن(سم) وعدد البذور/ نبات ونسبة البروتين الكلى عند مستوى معنوية أقل من 0.05. يستطيع البحث الحالي أن يوفر المعلومات من الصفات المورفولوجية والجزيئية من فول المونج، من المأمول أن تكون هذه النتائج مفيدة في تأكيد التراكيب الوراثية الجديدة، سيوفر موضع الحزم بواسطة SCoT صفات تحديد هذا النبات.

الكلمات الاسترشادية: الانتخاب الوراثي، فول المونج، أشعة جاما، الواسم الجزيئي SCoT.