*Egypt. J. Chem.* **Vol. 67**, No. 4 pp. 601 - 607 (2024)

**Egyptian Journal of Chemistry**

http://ejchem.journals.ekb.eg/

# Molecular Genetic Markers of Seven Lime and Lemon Cultivars

**Noran Ahmed[1] , Eman M. Fahmy[2] , Lamyaa M., Sayed[3] , Fahmy H.Abo Doma[4]**

[1] *Department of Genetics Faculty of Agriculture Ain Shams University*

[2] *Department of Genetics, Faculty of Agriculture, Ain Shams University, Kaliobeya, Egypt*

[3] *Genetics Dept., Fac. of Agric. Ain Shams University*

[4] *Genetic Department, Fac. Agric., Ain Shams University*

**Abstract**

Molecular genetic variability among seven limes and lemons cultivars was studied. The generated profiles revealed high levels of polymorphism with 79 fragments among the overall seven cultivars' using SCoT and ISSRs analyses (41 and 38, respectively). The data of Scot and ISSRs revealed total monomorphic bands of 43 (24 and 19, respectively). The total polymorphic fragments were 36 (14 and 22, respectively), with a polymorphism percentage of 53.65% - 36.64%, respectively. Moreover, the unique marker fragments were scored as 10 for SCoT and ISSRs analyses (5 for each one). The genetic distance (GD) of SCoT ranged from 0.86 to 0.972 and was higher than the range of the genetic distance of ISSRs which was from 0.750 to 0.933. At the same time, the genetic distance of the combined data was from 0.823 to 0.950. Cluster analysis by dendrogram tree divided the Lime and Lemon cultivars into two main clusters which was agreed with the origins of these cultivars. The outcome of this research will provide helpful and potential information for more studies in genetic diversity, population genetics, and genetic improvements in breeding programs of Lime and Lemon cultivars.

***Keywords***: Lime cultivars, Lemon cultivars, SCoT, ISSRs, Genetic diversity, Molecular distance.

## 1. Introduction

Limes are commonly propagated through seeds. So, there is a variation in some characteristics such as growth, fruit yield and quality, flowering seasonality, time of harvesting, and disease resistance through trees. To significantly improve the quality and productivity of limes, selection can play a vital role in this respect [14]. Several authors studied the variations between *citrus* trees to increase the number of genotypes involved in breeding programs and release new cultivars. It is hard to differentiate *citrus* cultivars using morphological traits because citrus trees and fruits usually do not bear until three to four years after planting. So apart from morphological characterization, it is desirable to develop alternative methods which are rapid, reliable, and more or less not influenced by the environment [21].

Morphological data in *citrus* taxonomy have been increasingly detected using molecular markers analysis. Moreover, molecular analysis data are highly efficient and introduce abundant information for environmental factors as well as molecular analysis have considered an important means for identifying genotypes and detecting of variations among different accessions and following the genetic inheritance of favorite economic traits. Molecular markers based on Polymerase Chain Reaction (PCR) such as SSRs, AFLP, ISSRs, and RAPD analyses are usually used in genetic taxonomy, genetic diversity, and identification between different species and cultivars [1,2, 3, 7, 9, 12, 18, 19].

Providing premium sources of genetic diversity assessment based on molecular genetic analysis are more important in breeding programs. Determining promising varieties can help breeders select characteristics economic character characteristics and increase economic crop productivity [13]. On the other side, this technique can produce dominant markers resulting from different sequences. The SCoT analysis was opinion to further additional dominant genetic markers analysis such as ISSRs and RAPD in higher polymorphism and in resolvability, which considered as a desirable marker [15], [17] in canolla [4] in El Amar apricot strains [6] in squash [20] in deciduous rootstocks [5] in apricot rootstocks.

This study investigated the genetic variation among the seven Egyptian Limes and Lemons cultivars by investigating the genetic variation among the seven Egyptian Limes and Lemons cultivars using two

*Corresponding author e-mail: nouranelmahalawy@gmail.com; (N. Ahmed)

Receive Date: 10 August 2023, Revise Date: 12 September 2023, Accept Date: 01 October 2023

DOI: https://doi.org/10.21608/ejchem.2023.228356.8404

©2024 National Information and Documentation Center (NIDOC)

different molecular genetic analyses (SCoT and ISSRs).

## 2 Materials and Methods

This study was done at the laboratories of the Genetics Department in the Faculty of Agriculture, Ain Shams University.

### 2.1 Plant materials

Leaves from seven cultivars of lime and lemon were sampled. Balady Rahmany, Balady Hosseiny, Sweet Lime, Shearer Lime, *and Banzaheir* Lime cultivars belong to *Citrus aurantifolia* and the Volka Lemon cultivar belongs to *Citrus Volkamariana* as well as, the Adalia Lemon cultivar belongs to *Citrus Limoul* of the *Citrus* genus were collected from two different orchards (Nawarit EL Hegaz 52K and El-Loaloah 54K) in Alexandria desert road of Egypt.

### 2.2 Molecular genetic markers

DNA was isolated from fresh tissue leaves for the seven cultivars of Lime and Lemon under investigation. Genomic DNA was used to generate molecular genetic markers using SCoT and ISSRs analyses.

### 2.3 DNA extraction

DNA was extracted from fresh tissue leaves using a mini kit for plants (bio basic) as described by the kit guide. Moreover, DNA quality was detected on 1% agarose gel electrophoresis using submarine gel BioRad at 100v for30m.

### 2.4 Polymerase chain reaction

DNA extracted from the seven lime and lemons cultivars were used as templates for six SCoT and six ISSRs primers based on polymerase chain reaction (PCR) analysis. Table (1) represents names and sequences of the used primers in molecular genetic identification among the seven Lime and Lemon cultivars. SCoT primers were designed according to a previous study by [11]. Amplified products of PCR were run on a 1.5% agarose gel electrophoresis and visualized using ethidium bromide and UV transilluminator. Molecular sizes were determined using a 100bp DNA Ladder marker (100bp to 3000bp). The running was done using mini submarine gel BioRad on 100 V for 30 min.

**Table 1**. SCoT and ISSRS primer names and their sequences for the analysis techniques

| SCoT | | | ISSRs | |
|---|---|---|---|---|
| P. no | Primer name | primer sequences (5/ ------ 3/) | Primer name | primer sequences (5/ ------ 3/) |
| 1 | SCoT 2 | ACC ATG GCT ACC ACC GGC | HB-8 | GAG AGA GAG AGA GG |
| 2 | SCoT 3 | ACG ACA TGG CGA CCC ACA | HB-9 | GTG TGT GTG TGT GC |
| 3 | SCoT 4 | ACC ATG GCT ACC ACC GCA | HB-10 | GAG AGA GAG AGA CC |
| 4 | SCoT 8 | ACA ATG GCT ACC ACT GAG | HB-11 | GTG TGT GTG TGT TGT CC |
| 5 | SCoT 10 | ACA ATG GCT ACC ACC AGC | HB-12 | CAC CAC CAC GC |
| 6 | SCoT 11 | ACA ATG GCT ACC ACT ACC | HB-13 | GAG GAG GAG GC |

### 2.5 Data analysis

Amplified DNA products were photographed and analyzed by GelAnalyzer3 software which scored clear fragments as present (1) or absent (0) for each primer and entered in the form of a binary data matrix according to [8].

According to the binary data matrix, genetic distances (GD) and cluster analysis were performed using the PAST 2 program and calculated according to Nei and Li coefficient.

## 3 Results and Discussions

### 3.1 Molecular genetic markers

Molecular genetic markers and genetic variability among the seven cultivars of Lime and Lemon under investigation were performed using SCoT and ISSRs analyses. Six SCoT and six ISSRs primers produced different banding patterns.

Amplified fragments of these two analyses were illustrated in Figs (1 and 2) and scored in Tables (2 and 3).

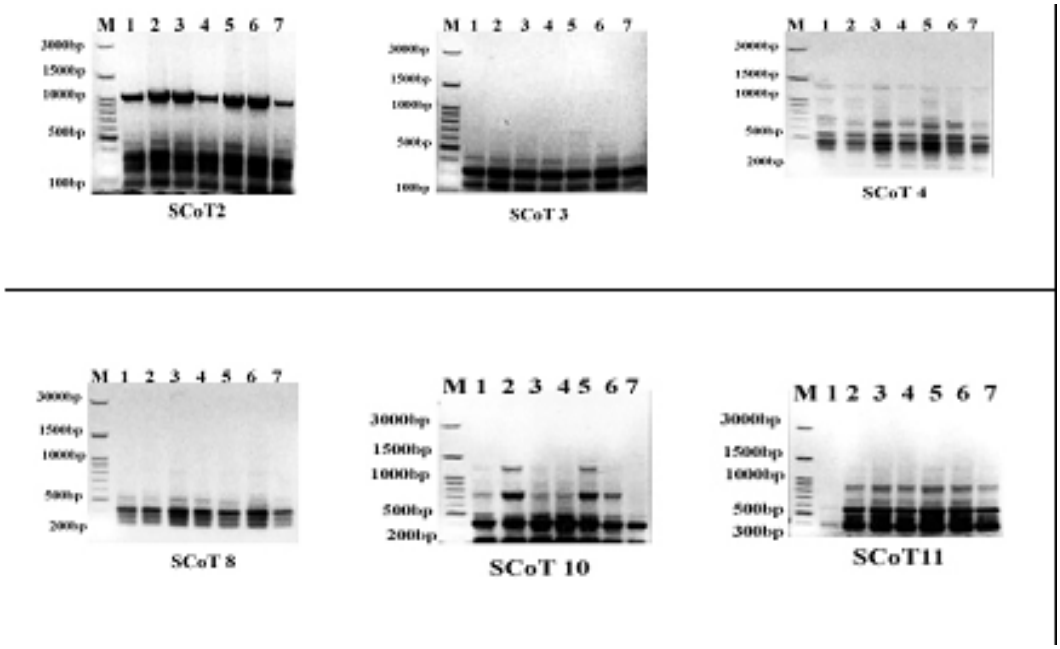


**Fig 1**. Amplified DNA fragments of SCoT -PCR products using six primers for the Seven Lime and Lemon cultivars; 1-Balady Rahmany 2-Balady Hosseiny 3- Sweet Lime 4- Sheaeer Lime, 5-Banzaheer Lime 6- Volka Lemon and 7-Adalia Lemon.

.

**Table 2.** Amplified DNA-PCR product data using the six SCoT primers with the seven cultivars.

| Primer name | M.W range (bp) | Total Frag | Monomorphic fragments | Polymorphic fragments | Unique markers | Polymorphism % |
|---|---|---|---|---|---|---|
| **SCoT 2** | **180 - 1080** | 8 | 6 | 2 | **1** | 25% |
| **SCoT 3** | **340 - 545** | 4 | 3 | 1 | **545bp (V. Lemon)** | 25% |
| **SCoT 4** | **200 - 1365** | 11 | 6 | 5 | **-** | 45.45% |
| **SCoT 8** | **200 - 825** | 5 | 4 | 1 | **-** | 20% |
| **SCoT 10** | **270 - 1385** | 5 | 3 | 2 | **745bp (A. Lemon)** | 40% |
| **SCoT 11** | **400 - 880** | 5 | 2 | 3 | **880,675,515bp (B. Rahmany)** | 60% |
| **Total** | | **38** | **24** | **14** | **5** | **36.84%** |

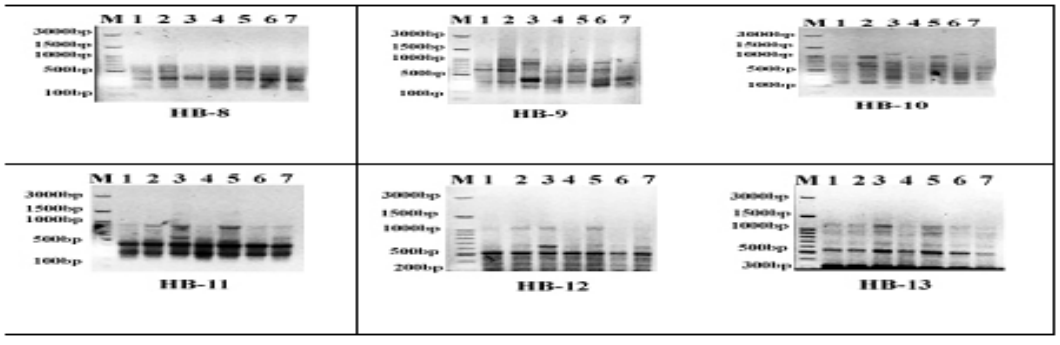


**Fig 2**. **Amplified DNA fragments product of ISSRs-PCR products using six primers for the seven Lime and Lemon cultivars:** 1-Balady Rahmany 2-Balady Hosseiny 3- Sweet Lime 4- Sheaeer Lime, 5-Banzaheer Lime 6-Volka Lemon 7-Adalia Lemon using six primers.

**Table 3.** Amplified DNA-PCR products data using the six ISSRs primers with the seven cultivars.

| Primer name | M.W range (bp) | Total frag | Monomorphic fragments | Polymorphic fragments | Unique markers | Polymorphism % |
|---|---|---|---|---|---|---|
| **Hb-8** | **220 - 700** | 5 | 1 | 4 | **220bp (S. Lime)** | 80% |
| **HB-9** | **270 - 1040** | 6 | 1 | 5 | **-** | 83.33% |
| **HB-10** | **240 - 1235** | 12 | 5 | 7 | **165bp (S. Lime) 760bp (S. Lime)** | 58.33% |
| **HB-11** | **280 - 900** | 6 | 3 | 3 | **-** | 50% |
| **HB-12** | **300 - 1080** | 6 | 5 | 1 | **1080bp (V. Lemon)** | 16.66% |
| **HB-13** | **335 - 1235** | 6 | 4 | 2 | **1000bp (S. Lime)** | 16.16% |
| **Total** | | **41** | **19** | **22** | **5** | **53.65%** |

**3.1.1 SCoT-PCR molecular genetic markers**

Overall SCoT primers' data for the seven Lime and Lemon cultivars were recorded as shown in Fig (1) and scored in Table (2). The results revealed that 38 amplified fragments were produced across the seven cultivars. Molecular size ranged from 180 to 1385 bp. The results showed 14 polymorphic and 24 monomorphic fragments with a polymorphism percentage of 36.84%. The highest polymorphism percentage produced with primer SCoT 11 was 60 %, while the lowest polymorphism percentage was 20% that appeared with primer SCoT 8. On the other hand, the number of amplified fragments for primer SCoT 4 (11 bands) was the highest one, while the number of amplified fragments (4) using primer SCoT 3 was the lowest one.

Unique markers resulted for the Balady Rahmany cultivar- which showed the best performance in productivity among the tested cultivars under this investigation. These unique markers were 3 negative specific-markers using primer SCoT 11 with molecular sizes of 515, 675, and 880 bp. While in cultivar Volka Lemon, a positive, unique marker with a molecular size of 550 bp appeared using primer SCoT 3. In the cultivar Adalia Lemon, a specific particular marker resulted in molecular size of 750bp using primer SCoT 10. These unique markers can be used for marker-assisted selection (MAS) in genetic improvement program in Lime and Lemon cultivars as revealed by [8] in Cymbopogon; [17] in EL Amar Apricot strains [2] in tomato [6] in squash and [20] in some deciduous rootstocks.

**3.1.2 ISSRs-PCR molecular genetic markers**

Molecular biodiversity was assessed among all the seven cultivars of Lime and Lemon under investigation using six ISSRs primers (Fig 2 and Table 3). The results revealed that the total number of fragments was 41, with molecular sizes ranging from 165 to 1235 bp. The obtained results showed a total of 22 polymorphic fragments with a polymorphic percentage of 53.65 %. The highest polymorphism percentage was 83.33% resulting from HB-9 primer, while the lowest polymorphism percentage was 16.66% and 16.16% which was presented using primers HB-12 and HB-13 respectively while primer, HB-10 was the highest in amplified fragments (12) while primer HB-8 was lowest in amplified fragments (5). On the other hand, 19 monomorphic fragments were produced among overall the six used primers.

Moreover, primer HB-10 produced a positive specific unique marker with a molecular size of 165bp in the Sweet Lime cultivar in addition, there were three negative specific unique markers made with the same cultivar using primers HB-8, Hb-10, and HB-13 with molecular weights (220, 760 and 1000bp), respectively. Moreover, the Volka Lemon cultivar produced one positive specific unique marker with a molecular size of 1080bp using primer HB-12 primer. These results confirmed that of [15] in Potato [17] in EL Amar Apricot strains [13] and in durum wheat [10 and 20] in deciduous rootstock.

**3.1.3 SCoT and ISSRs combined data analyses**

The data of the cultivars of Lime and Lemon for SCoT and ISSRs primers were combined (Table 4) that produced 79 fragments, where 43 out of them were monomorphic, and 36 were polymorphic bands with a polymorphism percentage of 45.56%. Seven unique SCoT markers were detected and could be used to tag cultivars that may be used in crop improvement programs [16].

Molecular fragments data estimated the genetic distance (GD) matrix among all the studied Lime and Lemon cultivars based on the combined SCoT results, and ISSRs fragments illustrated in Table (5). The GD based on SCoT fragments ranged from 0.862 between Balady Rahmany and Sweet Lime cultivars to 0.967 between Balady Hosseiny and Banzaheir Lime cultivars; and these cultivars are belong to *Citrus aurantifolia*, GD based on SCoT primers was higher than that found on ISSRs primers which were 0.750 between Balady Hosseiny and Adalia Lemon cultivars. At the same time, it was (0.933) between Balady

Rahmany and Balady Hosseiny cultivars. On the other hand, the genetic distance based on the combined data (0.823) was between Balady Rahmany and Adalia Lemon cultivars, and (0.950) was between Balady Hosseiny and Banzheir Lime cultivars which they belonged to *Citrus aurantifolia* From the previous results, SCoT analysis had more advantages in determining the genetic diversity of Lime and Lemon cultivars than the ISSRs analysis [5].

Fig (3) illustrates combined data of SCoT and ISSRs analyses, by dendrogram tree using the Dice-dissimilarity index according to [9]. The results showed that the dendrogram tree divided the cultivars into two main groups: the first leading group included two sub-groups; the first sub-main group included Balady Rahmany cultivar only, while the second sub-main group included Balady Hosseiny and Banzaheir Lime cultivars. On the other hand, the second main group was divided into two sub-main groups; the first sub-main group included Shearer Lime and Adalia lemon cultivars, while the second sub-main group included each of Sweet Lime and Volka Lemon cultivars.

These results of SCoT and ISSRs analysis for characterizing the genetic relationships among the Lime and Lemon cultivars were agreed with [17] in Apricot strains and [9] in some *Thymus* species. These clusters were conducted using ScoT and ISSRs primers results according to [6, 2, 10 and 5].

**Table 4.** Polymorphic, monomorphic, specific markers and Polymorphic percentage generated by the two techniques (SCoT and ISSRs) analysis.

| Primer name | Total Frag | Monomorphic fragments | Polymorphic fragments | Unique band | Polymorphism % |
|---|---|---|---|---|---|
| **SCoT** | 38 | 24 | 14 | 5 | 53.65% |
| **ISSRs** | 41 | 19 | 22 | 5 | 36.84% |
| **Total** | **79** | **43** | **36** | **10** | **45.56%** |

**Table 5.** Genetic similarity indices indices among the seven lime and lemon cultivars using the combined data of SCoT and ISSRs analysis A1(Balady Rahmany) A2(Balady Hosseiny) A3(Sweet Lime) A4 (Sheaeer Lime) A5(Banzaheer Lime) A6 (Volka Lemon) and A7(Adalia Lemon).

| MD | | A1 | A2 | A3 | A4 | A5 | A6 |
|---|---|---|---|---|---|---|---|
| A2 | ISSR | 0.933 | | | | | |
| | SCoT | 0.896 | | | | | |
| | Comb | 0.917 | | | | | |
| A3 | ISSR | 0.793 | 0.774 | | | | |
| | SCoT | 0.862 | 0.935 | | | | |
| | Comb | 0.836 | 0.864 | | | | |
| A4 | ISSR | 0.842 | 0.852 | 0.813 | | | |
| | SCoT | 0.892 | 0.933 | 0.966 | | | |
| | Comb | 0.859 | 0.886 | 0.902 | | | |
| A5 | ISSR | 0.847 | 0.920 | 0.754 | 0.866 | | |
| | SCoT | 0.896 | 0.967 | 0.967 | 0.966 | | |
| | Comb | 0.875 | 0.950 | 0.881 | 0.921 | | |
| A6 | ISSR | 0.777 | 0.758 | 0.857 | 0.836 | 0.842 | |
| | SCoT | 0.881 | 0.920 | 0.920 | 0.885 | 0.920 | |
| | Comb | 0.833 | 0.844 | 0.912 | 0.864 | 0.879 | |
| A7 | ISSR | 0.769 | 0.750 | 0.814 | 0.867 | 0.800 | 0.88 |
| | SCoT | 0.872 | 0.949 | 0.949 | 0.947 | 0.915 | 0.900 |
| | Comb | 0.823 | 0.854 | 0.888 | 0.914 | 0.854 | 0.886 |

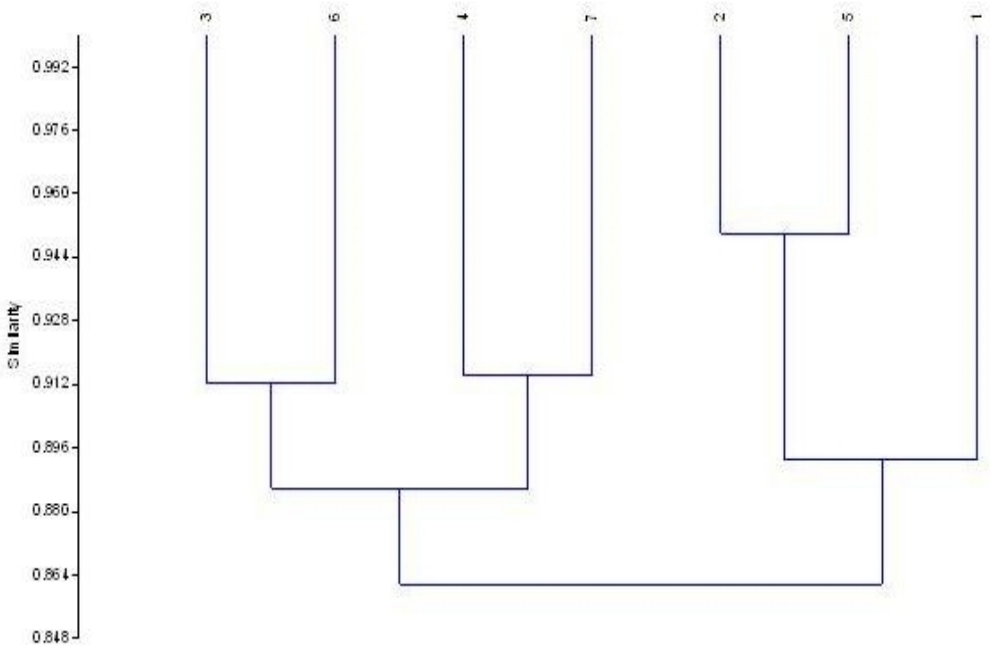


**Fig 3**. Hierarchical clustering of the similarity indices derived by UPGMA method using Dice-dissimilarity coefficient for combined binary data of SCoT and ISSRs analyses for the seven lime and lemon cultivars: 1-Balady Rahmany, 2-Balady Hosseiny, 3- Sweet Lime, 4- Sheaeer Lime, 5-Banzaheer Lime, 6- Volka Lemon, and 7-Adalia Lemon.

## 4 Conclusion

Generally, both SCoT and ISSRs analyses succeeded in generating reproducible and reliable DNA fragments. SCoT analysis was better than the ISSRs analysis in evaluating molecular genetic diversity that showed specific unique markers and more effective for assessing the genetic relationships among the studied lime and lemone cultivars. The outcomes of these results will offer helpful and potential information for more studies in population genetics, genetic diversity, and genetic improvements in Egyptian Lime and Lemon cultivar breeding programs.

## References

[1] Abdein M A E H (2018) Genetic diversity between Pumpkin accessions growing in the northern border region in Saudi Arabia based on biochemical and molecular Parameters. *Egyptian Journal of Botany* 58, 463-476. https://doi.org/10.21608/EJBO.2018.3612.1171

[2] Abdein M A E H, Abd El-Moneim D, Taha S S, et al (2018) Molecular characterization and genetic relationships among some tomato genotypes as revealed by ISSR and SCoT markers. *Egyptian Journal of Genetics and Cytology* 47, 139-159. https://journal.esg.net.eg/index.php/EJGC/article/view/281

[3] Abdein MA, El-Mansy AB, Awad NS, Abd El-Moneim D (2021) Assessment of genetic diversity in summer squash genotypes using some yield traits and DNA markers analysis under Sinai conditions. *Journal of Plant Production Sciences* 10, 13-29. https://doi.org/10.21608/jpps.2021.178522

[4] Abd El-Aziz M H, Farid S M, Elkomey S A A (2016) Evaluation of molecular and phenotypic diversity in relation to heterosis in some tomato lines under different climatic conditions. *Journal of Agricultural Chemistry and biotechnology* 7, 141-151. https://doi.org/10.21608/jacb.2016.40796

[5] Abd El-Aziz M H, Mohamed S Y, Magwaid H E (2019) Molecular and phytochemical assessment for some seedy strains of Alamar apricot rootstock under salinity stress. *Egyptian Journal of Basic and Applied Sciences* 6, 173-186. https://doi.org/10.1080/2314808X.2019.1690358

[6] Abd El-Hadi AH, Abd El-Aziz M H, Abd Alla MA, Ashak MG (2017) Molecular and phenotypic evaluation of some summer squash inbred lines. *Journal of Agricultural Chemistry and Biotechnology* 8, 281–287. https://doi.org/10.21608/jacb.2017.38913

[7] Abo El-Enien M M S, Kheder A M A (2018) Evaluation of a new thornless strain of Mexican Lime (*Citrus aurantifolia*, Swingle) Under Kaferelsheikh Governorate Conditions. *Egyptian Journal of Plant*

*Production* 9, 749-756. https://doi.org/10.21608/JPP.2018.36400

[8] Adhikari S, Saha S, Bandyopadhyay TK, Ghosh P (2015) Efficiency of ISSR marker for characterization of *Cymbopogon* germplasm and their suitability in molecular barcoding. *Plant Systematic and Evaluation* 301, 439-450. https://doi.org/10.1007/s00606-014-1084-y

[9] Alqahtani M M, Abdein M A, Abou El-Leel O F (2020) Morphological and molecular genetic assessment of some *Thymus* species. *Biosciences Biotechnology Research Asia* 17, 103-113. https://doi.org/10.13005/bbra/2815

[10] Baghizadeha A, Dehghanb E (2018) Efficacy of SCoT and ISSR markers in assessment of genetic diversity in some Iranian pistachio (*Pistacia vera* L.) cultivars. *Pistachio and Health Journal* 1, 37-43. https://doi.org/10.22123/PHJ.2017.54299

[11] Collard B C Y, Mackill D J (2009) Start Codon Targeted (SCoT) polymorphism: A simple novel DNA marker technique for generating gene-target markers in plants. *Plant Molecular Biology* 27, 86–93. https://doi.org/10.1007/s11105-008-0060-5

[12] El-Mansy AB, Abd El-Moneim D, Alshamrani SM, Alsafhi FA, Abdein MA, Ibrahim AA (2021). Genetic diversity analysis of tomato (*Solanum lycopersicum* L.) with morphological, cytological, and molecular markers under heat stress. *Horticulture* 7, 65 – 83. https://doi.org/10.3390/horticulturae7040065

[13] Etminan A, Pour-Aboughadareh A, Mohammadi R, Ahmadi-Rad A, Noori A, Mahdavian Z, Moradi Z (2016) Applicability of start codon targeted (SCoT) and inter-simple sequence repeat (ISSR) markers for genetic diversity analysis in durum wheat genotypes. *Biotechnology and Biotechnological Equipment* 30,1075-1081. https://doi.org/10.1080/13102818.2016.1228478

[14] Franck,C; Frederique, Ollitrault; Andres Garcia-Lor; Francois Luro; Luis Navarro and Pattck Ollitrault (2016) Phylogenetic origin of limes and lemons revealed by cytoplasmic and nulclear markers. *Annals of Botany* 117,565-583. https://doi.org/10.1093/aob/mcw005

[15] Gorji AM, Poczai P, Polgar Z, Taller J (2011) Efficiency of arbitrarily amplified dominant markers (SCOT, ISSR and RAPD) for diagnostic fingerprinting in tetraploid potato. *American Journal. Research* 88,226–237. https://doi.org/10.1007/s12230-011-9187-2

[16] Hajibarat Z, Said A, Hajibarat Z, (2015) Characterization of genetic diversity in chickpea using SSR markers, Start Codon Targeted Polymorphism (SCoT) and conserved DNA-derived polymorphism (CDDP). *Physiology and molecular biology of plants* 21,365–373. https://doi.org/10.1007/s12298-015-0306-2

[17] Mohamed S Y, Shoaib R M, Gadalla N O (2015) Selection of some seedling Apricot strains at Al-Amar region. *Journal of Applied Sciences* 15, 195-204. https://doi.org/10.3923/jas.2015.195.204

[18] Novelli V M, Machado M A, Lopes C R (2000) Isoenzymatic polymorphism in *Citrus* spp. and *P. trifoliata* (L.) Raf. (Rutaceae). *Genetics and Molecular Biology* 23,163-168. https://doi.org/10.1590/S1415-47572000000100030

[19] Osman A K E, Abdein M A E H (2019) Karyological and molecular studies between six species of Plantago in the Northern border region at Saudi Arabia. *Journal of Taibah University for Science* 13, 297-308. https://doi.org/10.1080/16583655.2019.1571400

[20] Safaa, M.S; Ehsan Nahla O, Mohamed S.Y and Thoria R.M (2018) Molecular genetic variability of some deciduous fruit rootstocks in Egypt. *Egyptian Journal of Scientific Research in Science* 35,95-111 https://doi.org/10.21608/jsrs.2018.26787

[21] Yadlod S S, Bhalerao R V, Pingle, S N (2018). Variability Studies of strains of kagzi lime (Citrus aurantifolia Swingle) in Latur district of Maharashtra. *Agricultural Science Digest* 38, 48-51. https://doi.org/10.18805/ag.D-4492