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# Identification of Four Molokhia (*Corchorus olitorius* L.) Genotypes by Molecular Markers

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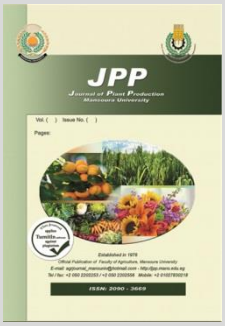


## ABSTRACT

Assessment of genetic diversity in a collection of genetic resources is a vital characterization for preserving this collection and planning future breeding programs. Genetic variability among four Egyptian molokhia (Corchorus olitorius L.) genotypes was observed using random amplified polymorphic (RAPD) DNA. RAPD patterns with polymorphism percentages were detected among the DNA samples from seven-day-old seedlings, representing each of the studied genotypes. Genetic diversity and relationships were evaluated using five randomly amplified polymorphic DNA markers that revealed 288 scorable bands. Amplification products were ranged in size from 110 bp to 5.5 kb, while in number, they ranged from 2 to 17 bands. Indicated significant genetic diversity among the molokhia genotypes were 66.67% for Wild type x Balady, 65.28% for Wild type x Saidy, 63.39% for Wild type x Falahy. The closest proximity, incidentally the highest genetic similarity, was observed between Balady and Falahy (80.56%), while the lowest was observed between Balady and Saidy (56.94%). The total positive unique bands scored 11 bands, while the negative unique bands scored 8 within the tested genotype and the five primers. Studied five primers generated unique bands in three molokhia landraces, which could identify these genotypes at the molecular level. The results suggest considerable potential RAPD approach for proper genetic identification of individual genotypes and an efficient way of varietal authenticity for fraud prevention. Genetic variability within molokhia genotypes ascertained using these primers. Results recommended the usefulness of RAPD primers in conservation and exploitation of molokhia germplasm in wide variety of breeding programs.

***Keywords:*** Molokhia, PCR, RAPD, Similarity, Unique bands, and Polymorphism.

## INTRODUCTION

The *Corchorus* genus includes 40-100 species of flowering plants in the Malvaceae family (formerly Tiliaceae), of which the most known cultivated species are *C. olitorius and C. capsularis* (Choudhary *et al*., 2013). Previously, the genus *Corchorus*, including *C. olitorius* and *C. capsularis* originated in Africa and later dispersed to Asia, however, the paraphyletic features of Tiliaceae transferred *Corchorus* and some other taxa to the subfamily Grewioideae, creating a monophyletic group within the family Malvaceae s.l. (Alverson *et al*., 1999; Bayer *et al*., 1999; Benor, 2018).

Molokhia is widely grown in the tropical and subtropical areas in the world, including Egypt as well as India, Bangladesh, China, Uzbekistan, Nepal, Vietnam, Burma, Zimbabwe, Thailand (Mir *et al*., 2008; Zhang *et al*., 2014 ; Zhang *et al*., 2015). Although molokhia genotypes are derived from the same pedigree, they may have different names in different places owing to the exchange of germplasm across different countries or regions. Thus, it is challenging to distinguish jute accessions using only morphological traits (Helaly *et al*., 2017).

*Corchorus* crops are available during the spring and summer seasons in Egypt and are considered excellent sources of vitamins (A, C, and E), proteins as well as mineral nutrients like calcium and iron (Dansi *et al*., 2008; Islam, 2013; Helaly *et al*., 2017). Moreover, *Corchorus olitorius* contains high iron and folate levels, which are useful for preventing anemia (Steyn et al., 2001). The molokhia plant, *C. olitorius* L., a recognized leafy vegetable and medicinal plant (Helaly *et al*., 2017), is rich in nutritional value with carotenoids, vitamins, phenolics, tannins, glycosides, mucilage, and minerals (Islam, 2013; Helaly *et al*., 2017). Furthermore, molokhia leaves and seeds have diverse phytochemical compounds such as cardiac glycosides, corchorin, corchorgenin, capsularin, corchoritin, olitoriside and corchortoxin (Ragasa *et al*., 2016; Hassan *et al*., 2019).

The richness of the molokhia plant with several healthy nutritional constituents offers many health benefits, such as the use of its leaves as an ingredient in a mucilaginous green soup in Egypt and are also reported to act as a demulcent, diuretic, and purgative (Oboh *et al*. 2012; Islam, 2013). Furthermore, in a recent report, Lee *et al*. (2020) reported the role of molokhia in preventing gut dysbiosis and obesity and its leaves is used for the treatment of pain, fever, and inflammation. Besides, the contents of certain phenolic and flavonoids compounds in molokhia provide great antioxidant, antiinflammatory, antitumor, antimicrobial, antiobesity, hypoglycemia, wound healing effects, and

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cytotoxic activities (Wang *et al*., 2011; Barku *et al*., 2013; Yan *et al*., 2013; Hassan *et al*., 2019).

Genetic diversity in the genus *Corchorus* is limited due to the existence of a robust sexual incompatibility between the two cultivated species, *C. capsularis* and *C. olitorius*, which prevents the development of improved cultivars. The limits of genetic characterization using morphological traits via field evaluation may be overcome by using the DNA molecular markers. Yet, crop identification has become increasingly important in the planning for breeding strategies and developing commercial interest in plant breeder rights and cultivar registration (Mady *et al*., 2013 and 2014; Helaly *et al*., 2014). However, closely related cultivars with low genetic variability cannot be readily distinguished by morphological indices (Degani *et al*., 1998; Singh *et al*., 2017).

Molecular markers are essential tools for measuring the diversity of plant species. Cost-effective assay, affordable hardware, high-throughput, convenience, and ease of test development and automation are essential factors when choosing a technology (Rafalski, 2002; Arif *et al*., 2010). Frequency data from markers such as random amplified polymorphic DNA (RAPD), inter simple sequence repeat (ISSR), amplified fragment length polymorphism (AFLP) and microsatellites provide the means to classify individuals into nominal genotypic categories and are mostly suitable for intra-species genotypic variation study. However, sequencing-based molecular techniques provide better resolution at intra-genus and above level (Robinson and Harris 1999). The variability of DNA-based techniques and the scope for characterization of genetic diversity in *Corchorus* sp. at the molecular level presents immense opportunities for developing new, improved cultivation.

Random amplified polymorphic DNA (RAPD) analysis on *Corchorus* cultivars has revealed polymorphism (Hossain et al., 2002). Besides, the RAPD-DNA technique was also used to assess the genetic diversity of many genotypes within plant species, such as summer squash (Mady *et al*., 2013); goldenseal (Inoue et al., 2013); and celery (Helaly *et al*., 2014). Genetic diversity of jute (*Corchorus* spp.) has been reported by Hossain *et al*. (2002) by using RAPD; Basu *et al*. (2004) using SSR and Roy *et al.* (2006) using STMS, ISSR and RAPD markers.

Furthermore, Hossain *et al*. (2003) characterized the cold-tolerant and cold-sensitive jute germplasms, and Qi et al. (2003) classified wild jute species using Inter Simple Sequence Repeat (ISSR) markers. Recently, Akter *et al*. (2008), Mir *et al*. (2008), and Huq *et al*. (2009) reported the usefulness of studying genetic variability for different traits in jute genotypes using jute-specific SSR markers. Moreover, molecular analysis via ISSR using five primers was done to identify heritability variation in scion and rootstock of grafted cucumber (Helaly *et al*., 2019).

A single RAPD primer generated six bands, revealed 50% total polymorphism, and divided the studied nine accessions of *C. capsularis* and *C. olitorius* into two clusters, one of five C. *capsularis* accessions and another cluster containing the remaining four accessions that belong to *C. olitorius* (Biswas *et al*., 2013). In a recent study, using five RAPD polymorphic primers, genetic diversity was assessed in 18 *C. olitorius* collected from different locations in Egypt and revealed a total number of 63 bands, of which 28 were polymorphic, representing 44.4% polymorphism. The authors observed that the RAPD primers used in the study generated between two and seven polymorphic bands (Youssef *et al*., 2019).

Considering the importance of *Corchorus olitorius* species as vegetable and fiber crops, with considerable commercial significance in several regions, so far, studies on genetic diversity and relatedness at the molecular level have been surprisingly scarce in molokhia genotypes or combined with other plant species. Using RAPD markers, this study investigated the genetic diversity and genetic relationships among the four collected Egyptian molokhia genotypes in different Governorates for information on molecular differentiation and characterization.

## MATERIALS AND METHODS

**Plant material.**

Wild type molokhia (*Corchorus olitorius* L.) seeds were collected and germinated in Petri dishes on Whatman paper, No.1 following previous studies by Helaly *et al*. (2008 and 2017). Genomic DNA was extracted from one whole seedling of each harvested genotype.

**Molecular analysis.**

Using Doyle and Doyle (1990) method, total genomic DNA was extracted from young seedlings of molokhia genotypes. Chemicals, random primers, and Taq polymerase kits were purchased from Sigma Aldrich, New England BioLabs, and Takara Co., USA. Genomic DNA fragments were amplified using five random RAPD primers with the sequences GGTAGCCGTC, CACACTGGCG, AGCGACCCGA, TTGGAGTGGC, ACCGACCTGT, and GTGTCCTGGC. The PCR reactions were done according to the manufacturer's instructions, using cycling protocol of 95°C for 3 min., 35 x (95°C for 50 sec., 37°C, 45 sec., 72°C, 1.2 min), and 72°C for 10 min.

The PCR bands were separated by electrophoresis at 50 volts on 1.5% agarose gel using one molar TBE buffer solution containing 6µL ethidium bromide. Molecular size was determined using a 100 DNA marker bp ladder. The bands were observed under UV-light to identify the polymorphic locus by size changes of DNA fragments. Polymorphism percentage was calculated using the following equation:

$$\text{Polymorphism } \% = \left(\frac{\text{No. of polymorphic amplified fragments}}{\text{Total No. of amplified fragments}}\right) \times 100$$

Bands were visually scored as either present (1) or absent (0). The unique bands that appear/disappear in a particular single genotype (defined as positive or negative unique bands as detailed in El-Khishin *et al*. (2003) and Helaly *et al*. (2014). Each separated band was interpreted as one allele, and the bands with the same mobility were assumed to be homologous (Van der Voort *et al*., 1997). Each marker was treated as an independent unit. In cases of similarity, the data were scored in the form of a binary matrix. For each pair of genotypes, the Jaccard similarity index (JS) was calculated (Jaccard, 1908).

The genomic DNA, extracted from seedlings of the investigated molokhia genotypes, served as templates for the PCR reaction, was surveyed using six RAPD primers, of which five were polymorphic.

## RESULTS AND DISCUSSION

### Results

Polymerase Chain Reaction (PCR) based technique using RAPD markers is now widely adopted in plant systematics and population biology to solve discrepancies in species identification, their classification as well as their hierarchical positions. The multi-locus fragments of RAPD as a dominant marker and the abundance throughout the genome along with the affordability and easiness of application, have made them very useful for identifying rare and endemic populations or accessions for conservation when other methods fail to detect variations or resolve relationships. Previously, Helaly *et al*. (2017a, b) identified the morphological and biochemical characteristics of the four studied molokhia genotypes.

In this study, the obtained genomic DNA amplification from each of the four genotypes using the five primers that produced polymorphic bands revealed various RAPD patterns (Fig. 1).

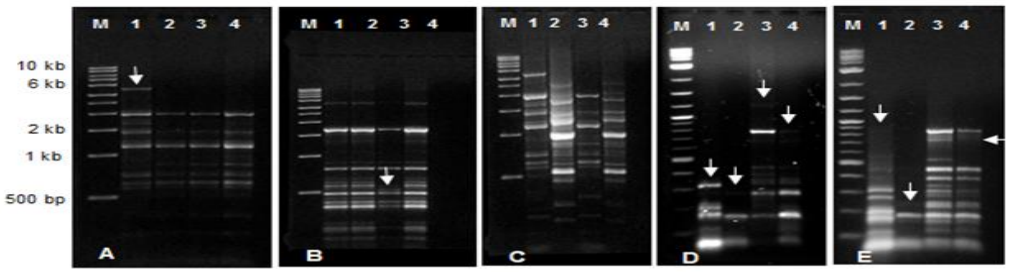


Figure 1. PCR amplification using random primers Co1 (A), Co2 (B), Co3 (C), Co4 (D) and Co5 (E). Molokhia wild type (lanes 1), Balady (lanes 2), Saidy (lanes 3) and Falahy (lanes4). Lane M contains a size marker (1 kb DNA ladder; Sigma Aldrich/Takara, USA).

The five primers produced 288 scorable bands, with 72 bands present and 216 absent bands. The number of bands produced by each primer varied. Depending on the use of the five RAPD primers and the four genotypes, the number of amplified bands scored 47, 43, 56, and 57 in Wild Type, Balady, Saidy, and Falahy genotypes, respectively (Table 1).

**Table 1. Number of the total scored bands, monomorphic bands, polymorphic bands and polymorphism percentage for each primer in four molokhia genotypes**

| Primers | Wild type | Balady | Saidy | Falahy | Total bands | Monomorphic bands | Polymorphic bands | Poly- Morphism% |
|---|---|---|---|---|---|---|---|---|
| Co 1 | 11 | 10 | 10 | 11 | 12 | 9 | 3 | 25.00 |
| Co 2 | 13 | 13 | 13 | 13 | 14 | 12 | 2 | 14.29 |
| Co 3 | 10 | 16 | 11 | 17 | 21 | 7 | 14 | 66.67 |
| Co 4 | 5 | 2 | 9 | 8 | 11 | 2 | 9 | 81.82 |
| Co 5 | 8 | 2 | 13 | 8 | 14 | 1 | 13 | 92.86 |
| Total | 47 | 43 | 56 | 57 | 72 | 31 | 41 | 56.13 |
| Mean | 9.4 | 8.6 | 11.2 | 11.4 | 14.4 | 6.2 | 8.2 | |

A total of 72 DNA bands were detected among the molokhia samples, with 31 (43%) being monomorphic and 41 (57%) being polymorphic. The five primers demonstrated genetic variability among the different molokhia accessions. The highest numbers of mono and polymorphic bands were 12 and 14 generated by Co2 and Co3 markers, respectively. On the other hand, the least scored reproducible fragments, 1 and 2, were obtained from Co5 and Co2 markers, respectively. The Co1, Co2, Co3, Co4, and Co5 markers revealed 25, 14, 67, 82 and 93% polymorphism, respectively, within the molokhia genotypes.

The maximum number of bands produced by the Co3 primer was 17 with Falahy genotype, while the minimum, two bands, were amplified by Co4 and Co5 primers with Balady genotype. The similarity interaction percentage (SIP) between primers and genotypes revealed that the molokhia wild type exhibited 68.6%, 62.8%, 64.8% similarity with Balady, Saidy and Falahy, respectively (Tables 2 and 3). Whereas the similarities between Balady, Saidy, and Falahy were 58.2 and 75.4%, and the similarity between Saidy and Falahy was 72.8%.

**Table 2. The similarity and dissimilarity percentage of DNA matrix in the various four molokhia genotypes**

| Genotypes | Similarity | | | |
|---|---|---|---|---|
| | Wild type | Balady | Saidy | Falahy |
| Wild type | 100 | 66.67 | 65.28 | 63.89 |
| Balady | 33.33 | 100 | 56.94 | 80.56 |
| Saidy | 34.72 | 43.06 | 100 | 70.83 |
| Falahy | 36.11 | 19.44 | 29.17 | 100 |
| | Dissimilarity | | | |

**Table 3. The scored number of bands of variant bands (SNVB) and similarity interaction percentage (SIP) among using four molokhia genotypes using five primers.**

| Primers | Total bands | Wild type X Balady | | Wild type X Saidy | | Wild type X Falahy | | Balady X Saidy | | Balady X Falahy | | Saidy X Falahy | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | SNVB | SIP | SNVB | SIP | SNVB | SIP | SNVB | SIP | SNVB | SIP | SNVB | SIP |
| Co 1 | 12 | 3 | 75 | 3 | 75 | 2 | 83 | 0 | 100 | 1 | 80 | 1 | 80 |
| Co 2 | 14 | 0 | 100 | 2 | 86 | 0 | 100 | 2 | 86 | 0 | 100 | 2 | 86 |
| Co 3 | 21 | 10 | 52 | 5 | 76 | 11 | 48 | 11 | 48 | 1 | 95 | 12 | 43 |
| Co 4 | 11 | 3 | 73 | 8 | 27 | 7 | 36 | 7 | 36 | 6 | 45 | 1 | 91 |
| Co 5 | 14 | 8 | 43 | 7 | 50 | 6 | 57 | 11 | 21 | 6 | 57 | 5 | 64 |
| Total | 72 | 24 | 343 | 25 | 314 | 26 | 324 | 31 | 291 | 14 | 377 | 21 | 364 |
| Mean | 14.4 | 4.8 | 68.6 | 5.0 | 62.8 | 5.2 | 64.8 | 6.2 | 58.2 | 2.8 | 75.4 | 4.2 | 72.8 |

**Identification of Molokhia genotypes.**

The four primers produced a total of 13 positive unique bands that are present in a particular genotype. Positive unique bands were distributed as five, seven, and one in wild type, Saidy and Falahy (Table 4).

The negative unique bands totaled eight, with two, four, and two in wild type, Balady, and Saidy, respectively. The genotype Saidy was characterized by the highest number of positive unique markers, six, in addition to two negative unique markers. Wild type was identified by five positive and two negative markers. Balady exhibited five negative unique markers and only one positive unique marker with Falahy, indicating that the different primers identified different numbers of unique markers. Positive unique bands scored 13 bands, while the negative unique bands scored 8 within each genotype and each primer. The five primers generated unique bands in three molokhia landraces, which could identify these genotypes at the molecular level. Primer Co5 exhibited the highest number of unique markers (10 bands); however, primer Co1 and Co2 showed the least unique bands, 2, among the studied genotypes.

The total number of unique markers per genotype ranged from 1 to 7 except Balady genotype recorded zero positive unique bands. The genotype Saidy was characterized by the highest number of unique positive markers (7), in addition to 2 unique negative markers. Wild type was identified by 5 positive and 1 negative markers. Falahy genotype exhibited one positive unique marker, while zero negative unique band. Data in Table 4 also illustrates that the different primer combinations identified different number of unique markers. Primer Co5 exhibited the highest number of unique markers, 4 positive and 4 negative. While, primers Co1 and Co2 were revealed the least one positive and one negative each.

**Table 4. The positive and negative unique band numbers and their size in four molokhia genotypes using five primers.**

| Primers | Positive unique bands | | | | Negative unique bands | | | |
|---|---|---|---|---|---|---|---|---|
| | Wild type | Balady | Saidy | Falahy | Wild type | Balady | Saidy | Falahy |
| Co 1 | 5.5 kb | - | - | - | 800 bp | - | - | - |
| Co 2 | - | - | 500 bp | - | - | - | 480 bp | - |
| Co 3 | 5.4 kb | - | 1.3 kb, 250 bp | 1.7 kb | - | - | 2.5kb | - |
| Co 4 | 120 bp<br>210 bp | - | 2 kb | - | - | 320 bp | - | - |
| Co 5 | 200 bp | - | 800 bp<br>750 bp<br>350 bp | - | 190 bp | 450 bp<br>310 bp<br>110 bp | - | - |
| Total | 5 | - | 7 | 1 | 2 | 4 | 2 | - |

**Discussion**

Genetic diversity using molecular marker can potentially anneal to homologous sequences in the entire genome, providing more excellent opportunities to uncover polymorphic regions and successfully employed in cultivar analysis among several plant species (Williams *et al*., 1990). After the variants of morphological and chemical characteristics of four molokhia genotypes, the RAPD data are more powerful in identifying those genotypes.

Several studies on some vegetable plants have routinely used morpho-agronomic traits as well as biochemical markers, including SDS-PAGE and isozymes, for varietal characterizations, i.e., cauliflower (Singh *et al*., 2021), celery (Helaly *et al*., 2014), squash (Mady *et al*., 2014 and 2020), and okra (Helaly et al., 2017a, b). Basically, the resolution of the molecular makers is much higher than morphological characters to identify individual varieties. In this study, the RAPD analysis results showed that molokhia genotypes could be identified by a certain number of RAPD markers amplified from a few primers. Therefore, DNA fingerprinting technology is an effective means for identifying molokhia genotypes (Islam *et al*., 2002; Islam *et al*., 2005) due to the discriminatory power of DNA-based molecular markers. Morphological attributes do not necessarily reflect real genetic relationships due to several limitations, such as the low level of polymorphism, low heritability, and late expression as well as the vulnerability to environmental effects (Smith and Smith, 1992; Konarev, 2000), and morphological data cannot provide reliable information on the calculation of genetic distance within the closely related genotypes.

In this study, the analysis of 41 polymorphic RAPD markers of 72 in this study suggesting a considerable genetic diversity among the included molokhia genotypes. Wide genetic variability among the tested genotypes suggests that these genotypes are highly diverse and suitable for involvement in breeding programs. The *Corchorus* spp. are self-pollinated and incompatible for inter-specific cross-hybridization, which results in a narrow genetic base. However, the high overall polymorphism percentage might be attributed to the distinctness of the tested genotypes, such as the black-seed wild genotype, which grows as a weed in cotton fields in Egypt.

Earlier studies used RAPD markers and revealed a lower polymorphism percentage among *C. olitorius* genotypes than the resulted percentage in this study, for example, 26% in Hossain et al. (2002), 28.3% in Heikal *et al*. (2015), and 44.44% in Youssef *et al*. (2019). The differences in the percentage of polymorphism among different studies are generally attributed to the number of primers and accessions used in the RAPD analysis. Similar results were found to attribute high polymorphism percentage within *Corchorus* spp. genotypes detected via RAPD, AFLP, and SSR (Martinello *et al*., 2001; Basu *et al*., 2004; Ur-rahman *et al*., 2004; Huq *et al*., 2009).

The positive and negative unique bands were essential for identifying molokhia genotypes from each other by their presence or absence. A positive unique band, present with a particular primer in a single genotype, but absent in the others, is known as a positive unique marker. However, the negative unique markers are absent in a specific genotype with a certain band. These bands are

useful for cultivar identification and fingerprinting due to their uniqueness and specific characteristics. RAPD analysis permitted the distinction among the molokhia genotypes and their characterization by specific unique markers. Moreover, each of the five primers (Co1, Co2, Co3, Co4, and Co5) could distinguish among the four genotypes.

The present results clearly demonstrate the efficiency of the RAPD marker system in molokhia genotypes fingerprinting using a few number of primer combinations. This could be attributed to the high multiplex ratio of the RAPD technique. These results are in consistence with the findings of several authors on *corchorus olitorius* genotypes (Hossain *et al*., 2002, Hossain *et al*., 2003; Mir *et al*., 2008; Heikal *et al*., 2015; Youssef *et al*., 2019) as well as many others on numerous plant species, such as celery, goldenseal, lettuce, squash, and okra (Inoue *et al*., 2013; Mady *et al*., 2013; Helaly *et al*., 2014; Helaly *et al*., 2017a, b).

## CONCLUSION

In the past, this approach had been successfully used to identify the four molokhia genotypes using five primers. RAPD technique can help validate genotype identity and address *ex-situ* conservation issues that involve varietal/genotypic identification. However, more accessions, species, and populations need to be analyzed before making generalized deductions on the genetic resources of molokhia populations. The highest similarity was observed between Balady and Falahy (80.56%), while the lowest was detected between Balady and Saidy (56.94%). These results indicated that molecular markers could be used for identifying genotypes, which is useful in planning and designing breeding programs for the improvement of molokhia germplasm.

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## تمييز أربعة تراكيب وراثيه من الملوخية (Corchorus olitorius L.) بواسطة الواسمات الجزيئية

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تحديد أربعة تراكيب وراثيه من الملوخية (Corchorus olitorius L.) عن طريق تقنية الواسمات الجزيئية الملخص العربى يعتبر تقييم التنوع الجيني لمجموعة من المصادر الوراثية توصيفًا حيويًا للحفاظ على هذه الأصناف والذى يفيد فى برامج التربية المستقبلية. لوحظ ان التباين الجيني بين الأربعة طرز الوراثية من الملوخية المصرية (*Corchorus olitorius* L.) باستخدام ال PCR وتقنية ال RAPD. تم الكشف عن أنماط ال RAPD بنسب تعدد الأشكال بين عينات DNAمن بادرات عمرها سبعة أيام ، تمثل كل من الطرز الوراثية المدروسة. تم تقييم التنوع والعلاقات الجينية باستخدام خمس بادئات عشوائية لل DNA والتي كشفت عن ٢٨٨ باند قابلاً للتسجيل. تراوحت نواتج ال PCR في الحجم من ١١٠ نيوكليوتيدة إلى ٥,٥ كيلو بايت ، بينما تراوحت في العدد من ٢ إلى ١٧ باند . وكان هناك اختلاف وراثي معنوي بين طرز الملوخية بلغ ٦٦,٦٧٪ للنوع البري x بلدي، ٦٥,٢٨٪ للنوع البري x صعيدي، ٦٣,٣٩٪ للنوع البري x فلاحي. ولوحظ ان درجة التقارب بين بلدي وفلاحي (٨٠,٥٦٪) ، وأقل درجة تشابه وراثي بين بلدي والصعيدي (٥٦,٩٤٪). سجل إجمالي الباندات الفريدة الإيجابية ١١ باند ، بينما سجلت الباندات الفريدة السلبية ٨ باندات للبادئات الخمسة. أنتجت البادئات الخمسة المدروسة نطاقات فريدة في ثلاثة سلالات من الملوخية ، والتي يمكن أن تحدد هذه الأنماط الجينية على المستوى الجزيئي. تشير النتائج إلى إمكانات كبيرة لنهج RAPD من أجل التحديد الجيني المناسب للأنماط الجينية الفردية وطريقة فعالة لمصداقية الأصناف لمنع الاختلاط. يمكن التحقق من التباين الجيني داخل الأنماط الجينية للملوخية باستخدام هذه البادئات. أوصت هذه النتائج بفائدة بادئات RAPDفي الحفاظ على الأصول الوراثية للملوخية واستغلالها في مجموعة متنوعة من برامج التربية.