RESEARCH ARTICLE

SABRAO Journal
of Breeding and Genetics
53 (3) 468-478, 2021

# ASSESSMENT OF ROOT-KNOT NEMATODE RESISTANCE IN EGGPLANT ACCESSIONS BY USING MOLECULAR MARKERS

**A. AINURRACHMAH[1,2], TARYONO[1,2*] and S. INDARTI[3]**

[1]Department of Agronomy, Universitas Gadjah Mada, Indonesia
[2]Agrotechnology Innovation Center, Universitas Gadjah Mada, Indonesia
[3]Department of Plant Protection, Universitas Gadjah Mada, Indonesia
*Corresponding author email: tariono60@gmail.com
Email addresses of coauthors: ainurrachmahaida@gmail.com, siwi.indarti@ugm.ac.id

## SUMMARY

Eggplant is one of horticultural crop species that is widespread in tropical and subtropical regions. Root-knot nematode (*Meloidogyne incognita*) is a biotic factor that is becoming a serious problem because it affects the eggplant root system and physiological activities, resulting in reduced eggplant growth and yield. This study was conducted to quantify eggplant resistance to nematodes in the field and to identify genetically resistant genes by using cleaved amplified polymorphic sequence markers. Results showed that the eggplant accessions that were classified as moderately resistant, namely *Solanum aculeatissimum* (SL-TE 74 and SL-TE 590) and *Solanum torvum* (SL-TE 589), were basically wild relatives and had fewer galls and female nematodes than other accessions. Both wild relatives have the potential to become donors for the development of nematode-resistant cultivars. The eggplant accessions SL-TE 44 and SL-TE 579 were identified as highly susceptible. Molecular testing with Mi-1 markers (Rex) revealed that all of the eggplant accessions produced 550 bp DNA fragments. The DNA fragments of SL-TE 74 and SL-TE 590 were cut into sizes of 550, 450, and 240 bp, whereas SL-TE 589 were divided into sizes of 550 and 240 bp. In the amplification by using the C8B marker, the amplification fragments of SL-TE 589 had sizes of 1500, 1100, and 500 bp, whereas SL-TE 74 and SL-TE 590 had sizes of 2000 and 900 bp. However, the susceptible eggplant accessions only produced one 700 bp DNA fragment.

**Keywords:** Root-knot nematodes, eggplant, cleaved amplified polymorphic sequence

**Key findings:** The Mi-1 and C8B markers can detect nematode-resistance genes in eggplant. *S. torvum* and *S. aculeatissimum* have the potential to become donors for producing nematode-resistant eggplant cultivars. The method of screening eggplant resistance to nematodes at the seedling phase is quite effective and can help accelerate the plant breeding process.

Manuscript received: April 13, 2021; Decision on manuscript: June 25, 2021; Accepted: July 22, 2021.


Communicating Editor: Dr. Samrin Gul

## INTRODUCTION

Eggplant is the most important vegetable in most tropical and subtropical countries (Rahman *et al*., 2011; Ismael *et al*., 2017) and is widely distributed around the world (FAOSTAT, 2015; Fita *et al*., 2015). In Asian and Mediterranean regions, eggplant is included among the top five most important vegetable crops due to its antioxidant, mineral and vitamin contents (Gramazio *et al*., 2015; Taher *et al*., 2017). The most important nutritional components of eggplant fruit are phenolic compounds, which are beneficial for a number of metabolic and cardiovascular ailments (Plazas *et al*., 2013).

Root-knot nematodes (*Meloydogyne incognita*), also known as Southern root-nematode, adversely affect eggplant production worldwide by inducing the formation of galls at their feeding sites and causing 16.67% losses in eggplant fruit yield (Zhou *et al*., 2018). Given that root-knot nematodes are soil-borne pathogens (Manzanilla-Lopez and Starr, 2009), their management is difficult (Ocal and Devran, 2019). Root-knot nematodes are dimorphic. In root-knot nematodes, the mouth is located at the anterior end and has a stylet. The stylet is extandable, and the nematodes can use it perforate the plant cell wall and then to suck out cell contents. Nematodes produces giant cells (gall) that inhibit the absorption and transport of water and nutrients in plants. *Meloidogyne* spp. has four juvenile stages in addition to the egg-laying phase of adult females. The L2 phase is the infective stage, and nematodes in this stage enter the roots near the root tips. When they reach the root meristem, the nematodes move into the differentiating vascular cylinder. At this stage, xylem parenchyma cells are selected for feeding structure development. These cells are then initiated upon the injection of salivary secretions from the dorsal esophageal gland. The cells become hypertrophic and increase, causing the roots to produce giant cells due to the association of *Meloidogyne* spp. These giant cells function as metabolic sinks that actively remove nutrients from the host plant for nematode development (Bleve-Zacheo *et al*., 2007).

Nematode resistance in crop plants can be classified as active or passive defense mechanisms (Huang, 2001). Preinfection resistance, a passive mechanism, occurs on the root surface or around the rhizosphere and can affect the penetration capability of nematodes. Root exudates can act as an attractant or repellent for nematodes. The resistance mechanism that is activated after nematode infection can affect physiological processes in the roots and include preventing nematode feeding and feeding site formation and inhibiting nematode development and nematode reproduction (Trudgill, 1991). If the constitutive defense mechanism is insufficient to protect the plant from pathogen infection, the pathogen will successfully enter plant tissue (Abd-Elgawad and Molinari, 2008).

Resistant cultivars are considered as the most effective, environmemtally safe, and ecofriendly approach (Devran *et al*., 2013). The investigation of resistant genes in different accessions plays an important role in improving crop plants through genetic resource conservation and utilization. Root-knot nematode resistance has been identified in some wild relatives of crop plants, such as tomatoes, yams, and chilies. The well-known Mi-1 gene controls resistance to three important nematode species and shares several structural motifs with other R genes, such as NBS and LRR, which are plant-based protein families that are required for resistance to viruses, bacteria, fungi, and nematodes (Zhou *et al*., 2018). Genetic resources can be evaluated morphologically in the seedling stage at the screenhouse, molecularly characterized by using DNA marker techniques, and further characterized morphologically in the field. This experiment developed a screening technique for evaluating eggplant accessions that were resistant to nematodes at the seedling stage. The selected resistant accessions were further

molecularly tested by using cleavage amplified polymorphic sequence (CAPS) DNA markers.

## MATERIALS AND METHODS

### Plant material

Twenty-four eggplant acessions were studied for root-knot nematode resistance at the seedling stage through field and molecular analyses. Among these accessions, 20 belonged to *Solanum melongena,* two land races belonged to *Solanum aculeatissimum*, and one each accession belonged to *Solanum ferox* and *Solanum torvum.* The accessions were collected from different regions of Indonesia and procured from Agrotechnology Innovation Centre, Universitas Gadjah Mada (AIC-UGM), Indonesia (Table 1). The field evaluation experiment was conducted at the greenhouse of AIC-UGM, and DNA extraction and molecular studies were carried out in the Laboratory at the Department of Agronomy, Faculty of Agriculture, Universitas Gadjah Mada, Indonesia.

### Eggplant accession assay for resistance to nematodes

Evaluation was carried out in a completely randomized design (CRD) with three replications. Nematode isolates were established as a single mass for pure cultures (*M*. *incognita*) and inoculated into eggplant seedlings at the two-true-leaf-stage or 3 weeks after germination. The eggplant seedlings were transplanted into 120 cm × 55 cm × 10 cm rectangular plant pot trays containing sterilized sand. One mL of nematode suspension containing 100 stage 2 juvenile was inoculated into the roots of each plant by three injection points.

The eggplant seedlings were uprooted 8 weeks after inoculation and evaluated in accordance with gall and reproduction indexes (Table 3). For the estimation of the nematode population in the roots, 0.5 g of roots from each replication was macerated with a sufficient amount of water in a Waring blender for 30 s, and the numbers of nematodes in various stages of development in the roots were calculated (Akhter and Khan, 2018).

### DNA extraction

DNA was extracted in five stages from 100 mg of fresh leaf tissue by using a Geneaid kit by Geneaid Biotech Ltd, Taiwan. The first stage was tissue destruction by crushing the sample. The second stage was lysis. At this stage, the crushed sample was added with Buffer GP1 and RNAse, then incubated at 60 °C for 10 min and mixed with buffer GP2. The third stage was the DNA binding stage. At this stage, the sample was mixed with a GP3 buffer. The fourth stage was washing. First, the sample was added with buffer. Then, a wash buffer was added. The fifth stage was DNA elution. At this stage, the sample was added with an elution buffer, then left to stand for 3–5 min to ensure that the elution buffer was completely absorbed. Finally, the samples were centrifuged at 14 000–16 000 × *g* for 30 s.

### CAPS marker analysis

Three types of CAPS primers were used to determine whether the tested eggplant accessions had genes that were related to resistance to root-knot nematodes (Table 2). The PCR conditions for resistance analysis against *M. incognita* were as follows: 180 s at 94 °C as the initial denaturation step, 60 s at 94 °C, 120 s at 55 °C, 120 s at 72 °C (30 cycles), 480 s at 72 °C, and a final cycle at 4 °C for an

**Table 1.** Eggplant accessions used in the study.

| No. | Eggplant accessions | Species | Province of origin in Indonesia |
|---|---|---|---|
| 1 | SL-TE 45 | *S. melongena* | Yogyakarta |
| 2 | SL-TE 579 | *S. melongena* | Yogyakarta |
| 3 | SL-TE 187 | *S. ferox* | West Borneo |
| 4 | SL-TE 28 | *S. melongena* | East Java |
| 5 | SL-TE 74 | *S. aculeatissimum* | Yogyakarta |
| 6 | SL-TE 580 | *S. melongena* | Yogyakarta |
| 7 | SL-TE 581 | *S. melongena* | Yogyakarta |
| 8 | SL-TE 582 | *S. melongena* | Yogyakarta |
| 9 | SL-TE 25 | *S. melongena* | Yogyakarta |
| 10 | SL-TE 589 | *S. torvum* | Yogyakarta |
| 11 | SL-TE 590 | *S. aculeatissimum* | Yogyakarta |
| 12 | SL-TE 13 | *S. melongena* | Yogyakarta |
| 13 | SL-TE 583 | *S. melongena* | Yogyakarta |
| 14 | SL-TE 18 | *S. melongena* | Yogyakarta |
| 15 | SL-TE 584 | *S. melongena* | Yogyakarta |
| 16 | SL-TE 42 | *S. melongena* | Yogyakarta |
| 17 | SL-TE 47 | *S. melongena* | Yogyakarta |
| 18 | SL-TE 15 | *S. melongena* | East Java |
| 19 | SL-TE 46 | *S. melongena* | Yogyakarta |
| 20 | SL-TE 44 | *S. melongena* | Yogyakarta |
| 21 | SL-TE 585 | *S. melongena* | East Java |
| 22 | SL-TE 586 | *S. melongena* | Yogyakarta |
| 23 | SL-TE 587 | *S. melongena* | Riau |
| 24 | SL-TE 588 | *S. melongena* | Riau |

**Table 2.** CAPS markers.

| Primer ID | Primer Sequence | Restriction Enzim | References |
|---|---|---|---|
| Mi-1 (Rex) | F: AACCGTGGACTTTGCTTTGACT<br>R: TAAGAACAGGGACTCAGAGGATGA | TaqI | Lee et al. (2015) |
| Mi-J | F: CTACGGAGGATGCAAATAGAA<br>R: AATCATTATTGTCACACTTCCCC | NtIII | Lee et al. (2015) |
| C8B | F:TACCCACGCCCCATCAATG | - | Reddy et al. (2016) |

**Table 3.** Root damage values in eggplant accessions caused by nematodes.

| Resistance Status | Gall Index |
|---|---|
| Immune | No galling |
| Highly Resistant | 1–2 gall/root |
| Very Resistant | 3–10 gall/root |
| Moderately Resistant | 11–30 gall/root |
| Susceptible | 31–100 gall/root |
| Highly Susceptible | >100 gall/root |

Source: Taylor and Sasser (1978)

indefinite period. The PCR amplification for CAPS was carried out in a 10 µl volume comprising 5 µl of master mix (2.25 mM MgCl, 0.2 mM dNTPs, 0.5 U Taq DNA polymerase, and 109 PCR buffer), 2 µl of DNA (25 ng/µl), 0.5 µl of primers, and 3.5 µl of nuclease-free water. The results of CAPS amplification were electrophoresed in 2% agarose gel in 1× TAE and visualized by staining with fluoresafe DNA. The DNA ladder size marker was used to estimate the size of the amplification results.

**Restriction enzyme digestion**

Given that electrophoresis produced monomorphic results, the analysis was continued by cutting the PCR-amplified DNA with the TaqI restriction endonuclease enzyme to identify the resulting band differences. The amplified pieces were cut with TaqI restriction endonuclease enzyme in a volume of 20 µl containing 15 µl of DNA obtained through CAPS amplification, 2 µl of 10× Buffer for TaqI, and 5 U TaqI. Cutting was followed by electrophoresis in 2% agarose gel. The gel was visualized as described above with the same transluminator.

**Data analysis**

Analysis of variance was carried out in accordance with CRD then continued with Bonferroni multiple comparison by using LSD test. Analysis was performed by using SAS 9.4 software. DNA fragment measurement was performed by using Gel Analyzer.

**RESULTS**

**Genotype screening for resistance to nematodes**

During screening at the seedling stage, root damage was scored in accordance with the guidelines of Taylor and Sasser (1978) (Table 3). Scoring revealed that the accessions SL-TE 74 and SL-TE 590 produced 33.12 and 31.52 galls, respectively (Table 4). However, the eggplant accessions SL-TE 74 and SL-TE 590 showed small galls, and their root systems functioned properly without root rot (Figure 1). The mean numbers of female nematodes found in the roots of SL-TE 74 and SL-TE 590 were also low and were aproximately 55.40 and 45.30, respectively. On the basis of these results, the genotypes SL-TE 74 and SL-TE 590 were classified as moderately resistant.

In the accession SL-TE 589, the average number of small galls formed was 16.96, whereas the mean number of female nematodes found in the roots was only 21.29. In addition, the roots were still functioning well. Given these results, the accession SL-TE 589 was classified as moderately resistant. Other accessions, i.e., SL-TE 187, SL-TE 580, SL-TE 45, SL-TE 25, SL-TE 46, SL-TE 583, SL-TE 13, SL-TE 582, SL-TE 588, SL-TE 42, SL-TE 47, and SL-TE 28, showed a large number of galls and female nematodes that ranged from 50 to 100. These accessions were considerend to be susceptible. In accessions SL-TE 579, SL-TE 581, SL-TE 585, SL-TE 587, SL-TE 44, SL-TE 586, SL-TE 18, SL-TE 15, and SL-TE 584, the average numbers of galls formed exceeded 100, and almost all the roots exhibited a large number of galls and were colonized, decaying, and functioning poorly. These accessions were placed in the highly susceptible category. In accordance with their root damage scores, the accessions SL-TE 74, SL-TE 590, and SL-TE 589 were identified as moderately resistant given its low number of galls and female nematodes. Overall, the eggplant accessions SL-TE 579 and SL-TE 44 were identified as highly susceptible, because they had the highest number of galls and female nematodes.

**PCR amplification**

Several types of markers have been designed to identify Mi-genes. Not all primers could produce DNA bands with

**Table 4.** Number of female nematodes and root knots in eggplant accessions.

| Eggplant accessions | Number of Galls | Number of female nematodes | Resistance Level |
|---|---|---|---|
| SL-TE 590 | 31.52 ± 0.23 d–e | 45.30 ± 0.21 bc | Moderately resistant |
| SL-TE 587 | 109.83 ± 0.21 ab | 107.96 ± 0.20 ab | Highly Susceptible |
| SL-TE 584 | 112.54 ± 0.21 ab | 83.78 ± 0.20 ab | Highly Susceptible |
| SL-TE 581 | 143.06 ± 0.21 a | 134.44 ± 0.19 a | Highly Susceptible |
| SL-TE 582 | 65.65 ± 0.21 a–d | 57.81 ± 0.20 abc | Susceptible |
| SL-TE 586 | 92.34 ± 0.21 ab | 108.37 ± 0.20 ab | Susceptible |
| SL-TE 580 | 75.19 ± 0.21 a–d | 81.48 ± 0.20 ab | Putative resistant |
| SL-TE 579 | 139.93 ± 0.21 a | 154.43 ± 0.19 a | Highly Susceptible |
| SL-TE 583 | 75.56 ± 0.21 a–d | 60.97 ± 0.20 abc | Susceptible |
| SL-TE 13 | 74.49 ± 0.21 a–d | 90.93 ± 0.20 ab | Susceptible |
| SL-TE 15 | 90.11 ± 0.21 ab | 95.43 ± 0.20 ab | Susceptible |
| SL-TE 18 | 93.06 ± 0.21 ab | 129.11 ± 0.19 ab | Susceptible |
| SL-TE 187 | 97.18 ± 0.21 ab | 85.69 ± 0.20 ab | Susceptible |
| SL-TE 25 | 78.66 ± 0.21 a–c | 79.14 ± 0.20 ab | Susceptible |
| SL-TE 28 | 50.10 ± 0.22 b–d | 91.79 ± 0.20 ab | Susceptible |
| SL-TE 42 | 62.32 ± 0.21 a–d | 72.16 ± 0.20 ab | Susceptible |
| SL-TE 44 | 105.9 ± 0.21 ab | 82.34 ± 0.20 ab | Highly Susceptible |
| SL-TE 45 | 75.94 ± 0.21 a–d | 88.80 ± 0.20 ab | Susceptible |
| SL-TE 46 | 75.33 ± 0.21 a–d | 74.82 ± 0.20 ab | Susceptible |
| SL-TE 47 | 59.19 ± 0.21 b–d | 82.26 ± 0.20 ab | Susceptible |
| SL-TE 74 | 33.12 ± 0.22 c–e | 55.40 ± 0.20 abc | Moderately resistant |
| SL-TE 589 | 16.96 ± 0.24 e | 21.29 ± 0.23 c | Moderately resistant |
| SL-TE 585 | 103.34 ± 0.21 ab | 103.36 ± 0.20 ab | Highly Susceptible |
| SL-TE 588 | 62.53 ± 0.21 a–d | 88.43 ± 0.20 ab | Putative resistant |
| CV (%) | 19.62 | 22.32 | - |

Means ± standard error with different letters indicate statistically difference at $P < 0.05$ on Bonferroni's LSD test. Resistance level based on the number of galls (Taylor and Sasser (1978).

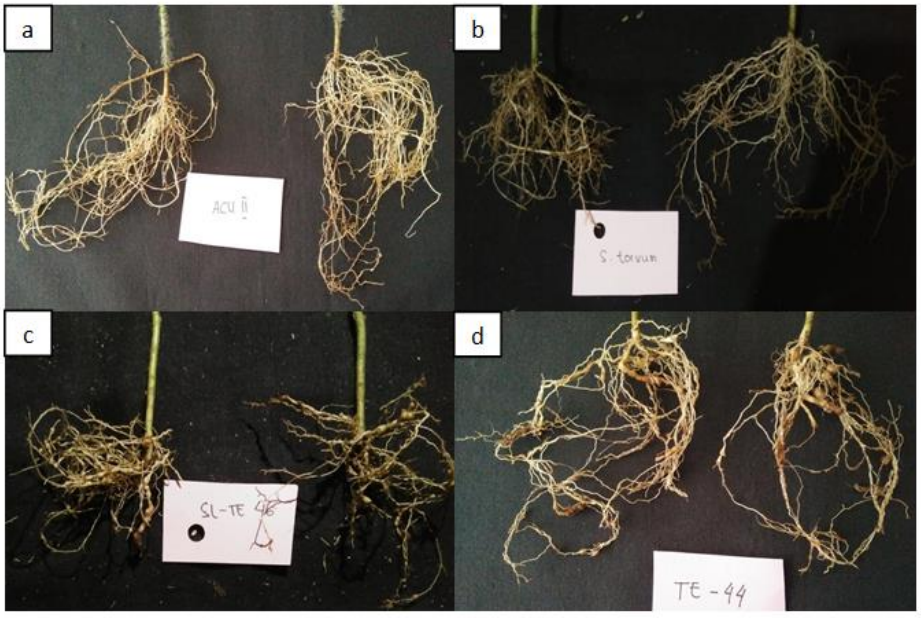


**Figure 1.** Root appearance on: a) moderately resistant (SL-TE 590), b) moderately resistant (SL-TE 589), c) susceptible (SL-TE 46), d) highly susceptible (SL-TE 44).

several CAPS markers, and the results for DNA amplification using Mi-J primers were unstable. In all eggplant accessions, the DNA amplification using the Mi-1 (Rex) marker produced similar DNA fragments with a size of 550 bp (Figure 2). After digestion with the TaqI restriction enzyme, the DNA fragment of SL-TE 590 and SL-TE 74 (moderately resistant) were cut into three bands, i.e., 550, 450, and 240 bp. In SL-TE 589 (moderately resistant), the DNA fragments was cut into two sizes, i.e., 550 and 240 bp, whereas the DNA fragments of susceptible eggplant genotypes were not restricted.

By using the C8B marker, most of eggplant accessions produced polymorphic DNA fragments even without digestion with the TaqI restriction enzyme. However, DNA fragments did not appear for all acessions during amplification by using the C8B marker. SL-TE 589 (Moderately resistant) showed 1500, 1100, and 500 bp amplification fragments (Figure 3). SL-TE 590 and SL-TE 74 (Moderately resistant) accessions had 2000 and 900 bp DNA fragments. However, the susceptible eggplant accessions produced only one DNA fragment with different band sizes for each accession.

A significant correlation was found between the Mi-1 and C8B markers and the level of root damage. The correlations of the Mi-1 marker with the numbers of galls and female nematodes were −0.605 and −0.0551, respectively, and those of the C8B marker with the numbers of gall and female nematodes were −0.416 and −0.469 (Table 5), respectively.

## DISCUSSION

Eggplant is classified as a perennial plant and can be grown in tropical and subtropical areas. Root-knot nematodes (*M. incognita*) adversely affect eggplant growth and result in failure to bear fruit. Resistance against pathogens only occur if the plant has a dominant resistant gene (R) that matches the dominant avirulent gene (Avr) of the pathogen. The Nem-R gene (resistant nematode) was first cloned from sugar beet Hs1pro-1, which can control sugar beet cyst nematodes (Cai *et al*., 1997). However, Hs1pro-1 has a weak association with the R gene of plants. Other Nem-R genes, which are cloned from tomatoes and potatoes, such as Mi-1, Hero A, Gpa1, and Gro1-4, are classified as the NBS-LRR gene R. The tomato genes Mi-1 and Hero A provide broad-spectrum resistance to several species of root-knot nematodes (Williamson and Kumar, 2006).

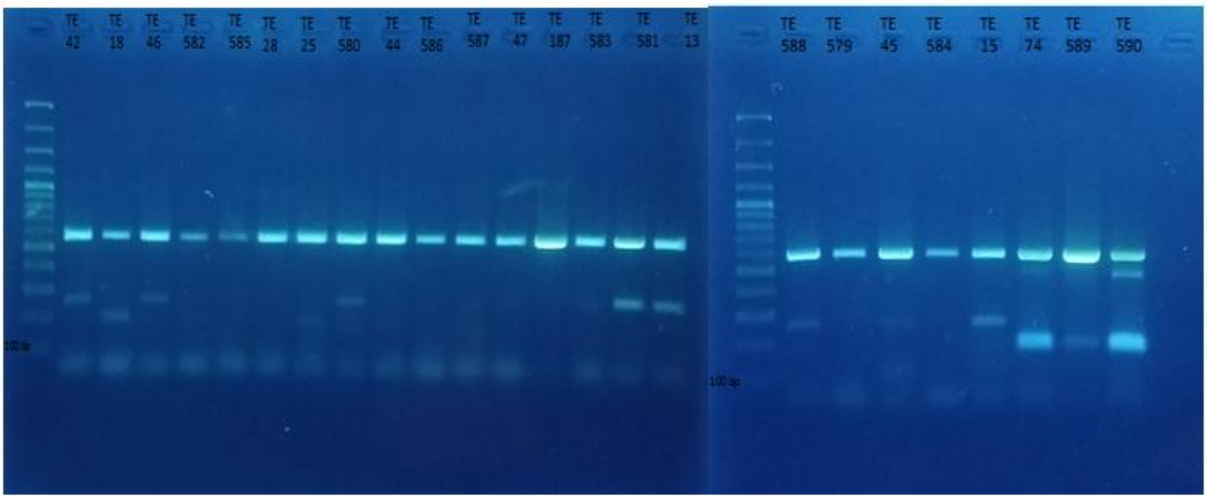


**Figure 2.** Amplification of the DNA CAPS marker Mi-1 in different eggplant accessions.

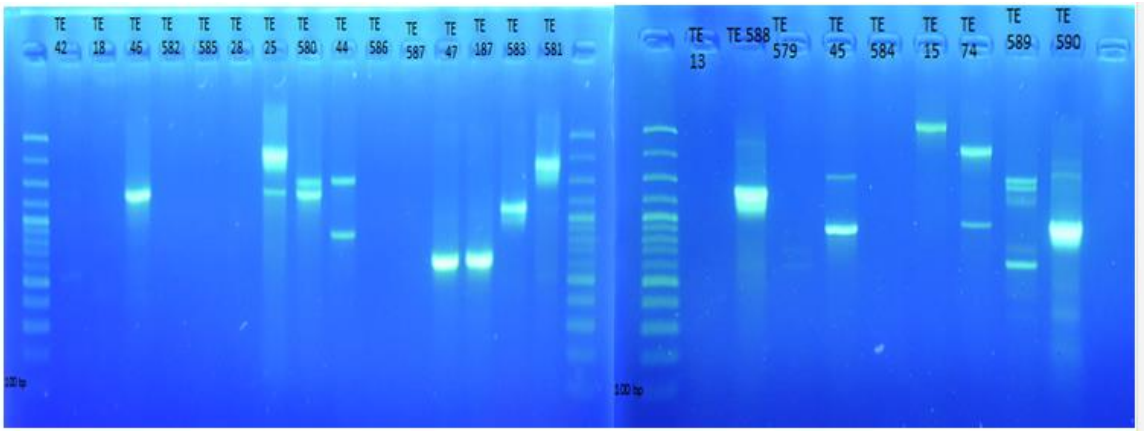


**Figure 3.** Amplification of the DNA CAPS marker C8B in different eggplant accessions.

**Table 5.** Correlation coefficients for Mi-1 and C8B markers with the level of root damage.

| Corr | Mi1 | Gall | Female |
|---|---|---|---|
| Mi1 | 1 | | |
| Gall | −0.605* | 1 | |
| Female | −0.551* | 0.8475* | 1 |
| Corr | C8B | Gall | Female |
| C8B | 1 | | |
| Gall | −0.416* | 1 | |
| Female | −0.469* | 0.848* | 1 |

Root-knot nematode resistance has been identified in crop plants and their wild relatives, such as the wild relatives of tomatoes, yams. and chilies. The Mi-1 gene has been widely recognized to show resistance to three important species of nematodes, namely, *M. incognita*, *Meloidogyne arenaria*, and *Meloidogyne javanica*. It can be isolated from wild tomato relatives by using the positional mapping approach and has been widely used in tomato cultivars. This single dominant gene shares several structural motifs with other R genes, including a nucleotide-binding site (NBS) and leucine-rich repeat domains (LRR), which are the features of plant-based protein families required for resistance against viruses, bacteria, fungi and nematodes (Zhou *et al*., 2018).

Juvenile nematodes infect susceptible and resistant plants. In susceptible plants, juveniles thrive in the roots, causing severe damage. The juvenile development of nematodes in the roots is reflected by the number of females that had formed by the end of the observation. In resistant genotypes, the nematodes are still capable of infection, and larvae can enter the roots in small numbers and develop into adults. In the most common resistance reaction, the larvae enter the roots, but cells from the larva die, causing the localized necrosis of plant cells around the anterior end of the nematode (Reddy *et al*., 2018).

The nematode resistance shown by the SL-TE 74, SL-TE 590, and SL-TE 589 accessions is the novel finding of the this study. Several eggplant accessions that were screened for resistance to nematodes by using the PCR method produced DNA amplification bands. The field screening test of eggplant accessions revealed that SL-TE 74 and SL-TE 590 were slightly resistant genotypes. The molecular analysis of all accessions with Mi-1 markers (Rex) produced DNA

fragments with a size of 550 bp. After digestion with the TaqI enzyme, the DNA fragments of the SL-TE 74 and SL-TE 590 accessions were cut into 550, 450, and 240 bp fragments. Amplification with the C8B markers produced DNA fragments with sizes of 2000 and 900 bp.

The field screening test showed that the eggplant accession SL-TE 589 was classified as moderately resistant to nematodes. This eggplant genotype produced DNA fragments of 550 and 240 bp after molecular analysis with Mi-1 markers (Rex) and digestion with the TaqI enzyme and produced DNA fragments with sizes of 1500, 1100, and 500 bp after evaluation with the C8B marker. Past findings showed that tomato DNA amplification with the CAPS Rex-1 marker produced DNA bands with a size of 750 bp in all genotypes (Reddy *et al*., 2016). After digestion with the TaqI enzyme, the resistant genotype was divided into two fragments with sizes of 570 and 162 bp. However, the fragments of the susceptible genotype could not be digested.

In this study, apart from SL-TE 590, SL-TE 589, and SL-TE 74, there were other accessions namely SL-TE 580 and SL-TE 588 (*Solanum melongena)* having the potential as resistant gene donor plants. The response of these two accessions to nematode attack were not severe. The gall number that was formed were 75.19±0.21 and 62.53±0.21, while the mean number of female nematodes found in roots were 81.48±0.20 and 88.43±0.20, with roots still well-functioning. In the DNA amplification step using the Mi-1 (Rex) marker after being digested using the TaqI restriction enzyme, the DNA fragment of SL-TE 580 and 588 were cut into two sizes i.e., 550, and 240 bp. Meanwhile by using the C8B marker, the amplification fragments of SL-TE 580 appeared at 1500 and 1100 bp and of SL-TE 588 appeared at 2000 and 1500 bp. In further research, it is necessary to carry out further evaluation to the two accessions to determine their potential as donor plants.

Molecular analysis supported morphological screening technique for nematode resistance. This finding was similar to the result of Soya and Tanyolac (2010) for tomato, where DNA amplification with CAPS Rex-1 marker provided similar DNA bands with sizes of 750 bp. After digestion with the TaqI restriction enzyme, the DNA fragment of the homozygote resistant genotype was cut into 570 and 160 bp bands, whereas the DNA fragments of the heterozygote resistant genotypes were cut into 750, 570, and 160 bp bands. The TaqI restriction enzyme was unable to cut DNA fragment of susceptible genotypes,

Root-knot nematodes severely affect eggplant growth and result in failure to bear fruit. Therefore, breeding to develop high-yielding cultivars with resistance to root-knot nematodes is very important. However, such cultivars have not yet been successfully produced. In this experiment, three candidates for resistance gene sources were found in wild relatives, namely SL-TE 74, 589, and 590. Transferring resistance genes from wild eggplant relatives into cultivated eggplant is necessary. Successful interspecific hibridization between *S. aculeatissimum* and *S. melongena* provides the opportunity to utilize resistance genes from *S. aculeatissimum* (Zhou *et al*., 2018). In the future, prebreeding is needed to evaluate the effectiveness of *S. torvum* and *S. aculeatissimum* as parental donors.

## CONCLUSIONS

Field evaluation revealed that SL-TE 74, SL-TE 590, and SL-TE 589, the wild relatives of eggplant, were donors of resistance to *M. incognita*. The Mi-1 (Rex) and C8B CAPS marker, which were tightly linked with the Mi gene, were found in eggplant. The DNA fragment of SL-TE 74 and SL-TE 590 (moderately resistant), were cut into three bands with sizes of 550, 450, and 240 bp. The DNA fragments

of SL-TE 589 (moderately resistant), was cut into two bands with sizes of 550 and 240 bp. By contrast, the DNA fragments of susceptible eggplant genotypes could not be restricted. The C8B CAPS marker has the advantage over the Mi-1 marker because the restriction step is not required. The three genotypes identified here have the potential to become donors for the assembly of nematode-resistant eggplant cultivars.

## ACKNOWLEDGEMENTS

The author would like to gratefully acknowledge Agrotechnology Innovation Centre -Universitas Gadjah Mada, Indonesia for providing eggplant breeding material. This research was funded by Program Penelitian Kompetitf Nasional Category Penelitian Dasar by Direktorat Riset dan Pengabdian Masyarakat Direktorat Jenderal Riset dan Pengembangan Kementerian Riset, Teknologi, dan Pendidikan Tinggi, Indonesia.

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