

POSSIBILITY OF COMBINING HIGH YIELD AND RESISTANCE TO *FUSARIUM* WILT DISEASE USING MOLECULAR MARKERS IN 4 ÉLITE SESAME LINES

Ayman Saber ANTER* and Ghada M. SAMAHA

National Research Centre, Field Crops Research Department, Dokki, Giza, EGYPT.

* Corresponding author: tokaeman@gmail.com

Received: 03.02.2020

ABSTRACT

Developing a high yielding variety connected with stress-resistant in sesame is a viable option to address the adverse effects of climate change. The objectives of this study were to identify high-yielding and to detect some molecular markers associated with *Fusarium* wilt resistance in sesame. Five genotypes were evaluated based on seed yield ha⁻¹ over three growing seasons (2016-2018) at two sites, Al-Nubaria (2016-2018) and Abu-Hammad (2016) in Egypt. Twenty RAPD and five ISSR primers used to detect some markers linked to *Fusarium* wilt resistance. Genotypes and environments and interaction between them showed high significant variation ($p < 0.05$) for seed yield ha⁻¹. The mean performance of the lines C1.5, C3.8, C6.3, and C1.6, for seed yield ha⁻¹ were higher than check variety by 3.4, 2.8, 0.5 and, 16.7%. Line C1.6 achieved less value of the standard deviation of ranks, based on seed yield ha⁻¹, through environments, indicating that it was less affected by environmental conditions. Molecular marker analysis revealed eight markers linked to *Fusarium* wilt resistance, they are seven positive markers (five RAPD and two ISSR) which were found in the line C3.8 and absent in the check variety. Finally, both C1.6 and C3.8 offering prospects to form new varieties sesame having high-yield and *Fusarium* wilt disease resistance.

Keywords: Climatic change, *Fusarium* wilt, molecular marker, *sesamum indicum*.

INTRODUCTION

Climate changes may contribute to biotic and abiotic stresses, which have negative impacts on global agricultural production, and the agricultural process is based on three main axes, the pathogen, and the host and the interaction with environmental conditions, where the relationships between them is the main key to the manifestation of infection, where climate changes has, among these factors, the main impact and these changes cause a decline in crop productivity (Prasch and Sonnewald, 2013; Ramegowda and Senthil-Kumar, 2015; Pandey et al., 2017; Raza et al., 2019). Plant breeding is an ideal method for facing the severe effects posed by the climate changes by creating new varieties that combine high yield and diseases resistant (Salme and Cagirgan, 2010; Lenaerts et al., 2019). Moreover, the ability of breeders to identify and access new varieties are becoming increasingly important as long as climate change continues.

In Egypt, sesame (*Sesamum indicum* L.) crop has high potential as a promising crop suitable for dry areas because it needs low water requirements, adaptation to soil and weather conditions, and is also well suited to

replace low-yield crops, especially under climate changes (Boureima et al., 2011; Misganaw et al., 2015; Dossa et al., 2017) and the sesame oil, other than its use as edible oil medium has specific industrial uses, make hair oil, hydrogenated oil, as well as a number of medicines. In addition, sesame seed is a good source of phosphorus, iron, magnesium, manganese, zinc and is rich in vitamin B (Suja et al., 2004; Orruno and Morgan, 2007; Quasem et al., 2009). Also, sesame may appreciably contribute to increasing the income of small farmers and plays an important role in accessing food for a lot of people by access to food and nutrition (Anilakumar et al., 2010; Baraki and Berhe, 2019). However, productivity in Egypt has been almost stagnant for a long time and *Fusarium oxysporum* sesame (FOS) is one of the most critical restrictions on the extension of cultivated area sesame in Egypt. This disease-causing quantitative and qualitative losses of yield (Khalifa, 2003; El-Shakhess and Khalifa, 2007; Shabana et al., 2014). Certainly, the development of high-yielding and disease resistant sesame varieties can offer permanent control over plant diseases and a low-cost and a more favorable environment and thus may partly contribute to solving the problem of oil production in Egypt. In addition, identifying the superior

line is a key part of the plant breeding process (Boureima and Abdoua, 2019).

The combination of modern technologies in classic plant breeding programs can much change the agricultural productivity, so the practical use of the molecular marker is a significant for accurately identifying markers associated with the target trait (Kumar and Sharma, 2011; Ahmad et al., 2017; Cobb et al., 2019). And many studies of linkage analysis of wilt resistance genes showed that the resistance genes and the markers were located in at the same linkage group, they tend to stay together as each generation of plants is produced. RAPD, ISSR could be helpful for the identification of resistant genotypes and effective utilization in marker-assisted selection if it is validated on different genetic backgrounds (Maisuria et al., 2017). Many previous studies used RAPD and ISSR marker to identify markers linked to *Fusarium* wilt

resistance in chickpea (Ratnaparkhe et al., 1998 a, b) and in rice and wheat (Prabhu et al., 2009). The objectives of this study were to identify high-yielding and to detect some molecular markers associated with *Fusarium* wilt resistance in elite lines of sesame.

MATERIALS AND METHODS

Plant material and experimental design

Breeding materials used in this investigation were 4 elite derived lines of sesame (F₈ to F₁₀ generations) namely: C1.5, C1.6, C3.8 and C6.3. And commercial variety Shandaweel (C). These materials obtained via pedigree selection from a continuous breeding program initiated at the Agronomy Department, Faculty of Agriculture, Cairo University while C brought of ministry agricultural. The characterizes of their parents and C presented in Table 1.

Table 1. The origin, breeding status and description for parents of lines and C.

| Genotypes | Breeding status | Seed source* | Specific characters |
|----------------|----------------------------|--|--|
| P1 (HM19) | F ₈ -hybrid pop | Cairo Univ.* | Early maturity, non branching, first capsule set low, 3 capsules/axil, high resistant against <i>Fusarium Oxysporum</i> . |
| P2 (EUL90) | Mutant line | Cairo Univ.* | Early maturity, non-branching, first capsule set low, 3 capsule/axil, moderate resistant against <i>Fusarium Oxysporum</i> . |
| P3 (Mutant 48) | Mutant line | Cairo Univ.* | Branching, 3 capsules/axil. high susceptible against <i>Fusarium Oxysporum</i> . |
| P4 (Giza 32) | Local variety | Ministry of Agric.& Land Reclamation, Egypt | Heavy seed weight, medium branching, one capsule/axil, long capsule, late maturity, moderate resistant against <i>Fusarium Oxysporum</i> . |
| P5 (NM59) | Exotic line | India through IAEA** | Stiff stem, late maturity, one capsule/axil, resistant against <i>Fusarium Oxysporum</i> . |
| P6 (Babil) | Exotic variety | Iraq through IAEA** | Low branching, 3 capsules/axil, semi-shattering capsules, resistant against <i>Fusarium Oxysporum</i> . |
| C | Local variety | Ministry of Agric. & Land Reclamation, Egypt | Heavy seed weight, medium branching, three capsule/axil, long capsule. susceptible against <i>Fusarium Oxysporum</i> . |

* Advanced breeding materials resulted from the breeding program conducted at Agron. Dept. Fac. of Agric. Cairo Univ. ** International Atomic Energy Agency.

This study was complementary to another study conducted by Shabana et al.,(2014).

They were tested for their reaction against infection with either *Macrophomina phaseolina*, *Fusarium oxysporum* or their combination. Fungal inoculation of *M. phaseolina* and *F. oxysporum* were prepared using sorghum-coarse sand-water (2:1:2 v/v) medium. The ingredients were mixed, bottled and autoclaved for two hours at 1.5 air pressure. The autoclaved media in glass bottles was inoculated separately using agar discs obtained from the periphery of a 5-day old colony of each of the tested fungi and incubated at 26°C for two weeks. They were then used for soil infestation. Each fungal inoculum's was added separately or in their mixture to the sterilized potted soil at the rate of 2% by weight, mixed thoroughly with the soil surface, then watered and left for one week

before sowing. Disease assessment was measured as percentages of pre- and post-emergence damping-off after 15 and 45 days from sowing, respectively. Percentages of diseased plants infected with charcoal-rot, wilt or charcoal-rot & wilt were estimated according to a specific disease symptoms and recorded after 90 days from sowing. Charcoal-rot infection was expressed as root discoloration, black stem rot and pronounced reduction in root system of the infected plants. However, infected plants were characterized by the internal vesicular discoloration wilt appearance and might be died and fell down and then they considered wilted. They found that P₅ and F₆ derived lines (C1.6, C3.8 and C6.3) were the most resistant lines against *Fusarium oxysporum* infection under greenhouse conditions while current study aimed to determine resistance lines based on the level genetics.

Trials were carried out during three growing seasons (2016-2018) in two locations, first at Agricultural Production and Research Station at National Research Centre, Al-Nubaria district, El-Behera Governorate in sandy soil and sprinkler irrigation applied. Second, at Arab Zaidan Village-Abu Hammad district, AL-Sharqia Governorate for one year (2016) in clay soil (Table 2) and irrigation surface applied. Sowing dates during three

seasons as follows: May 1, 10 and 13 at the Al-Nubaria site and May 8 at the AL-Sharqia site. Genotypes arranged in randomized complete blocks design with three replicates. Plots consisted of three rows, 3 m long and spaced 0.50 m apart with a 10 cm plant distance. Evaluation based on seed yield ha⁻¹: the total seed yield (kg/ha⁻¹) harvested from the net plot area (4.5m²). The recommendation of the Agricultural Ministry applied.

Table 2. Mechanical and chemical properties of the experimental soil in Al-Nubaria and AL-Sharqia sites.

| Mechanical properties | Unit | Al-Nubaria | AL-Sharqia |
|------------------------------|-------------|-------------------|-------------------|
| Coarse | % | - | 3.5 |
| Sand | % | 92.0 | 12.5 |
| Silt | % | 3.0 | 36.4 |
| Clay | % | 5.0 | 47.6 |
| Chemical properties | | | |
| PH | - | 7.6 | 8.03 |
| CaCO ₃ | % | 1.42 | 28.7 |
| EC | ds/m | 0.36 | 0.28 |
| Organic matter | % | 0.38 | 1.85 |

ds/m:Decisiemens/m

Statistical analysis

Combined analysis of variance, standard deviation (SD), and coefficient of variability (CV%) computed by program MSTAT-C (MSTAT-C, 1991).

Molecular analysis

The lines were classified as highly resistant to *Fusarium* wilt at seedling and mature stages except line C1.5 (Shabana et al.10). So, according to their resistance, the C3.8 line was chosen as the highest resistant one, while C was chosen as the susceptible one to *Fusarium* wilt.

Genomic DNA extraction

Genomic DNA was extracted from genotypes using DNeasy Plant Mini Kit (Qiagen) according to the manufacturer's instructions. The DNA isolated was checked on a 1 % agarose gel.

RAPD and ISSR analysis

A total of 25 primers (20 RAPD and 5 ISSR) were used in PCR amplification were synthesized by Sigma Aldrich, India. PCR reactions were performed according to (Williams et al., 1990) in DNA thermal cycler. The reaction mixture (25 µl) contained a 2.5 µl of 10× PCR buffer, 0.5 µl of dNTPs (2.5 mM), a 2.5 µl of MgCl₂ (2.5 mM), 2.0 µl of primer (RAPD and ISSR, respectively), 1U of Taq DNA polymerase enzyme (Fermentas) and 2 µl of DNA template. The PCR reactions were performed for both methods under following conditions: initial denaturation at 94 °C for 4min; 35 cycles of 1min denaturation at 94 °C; 1min annealing at a temperature specific to the primer and extension primer at 72 °C for 2min and 10min at 72 °C for

the final product extension. The amplification product was separated by electrophoresis on agarose (1.5%) in 1× TBE buffer run for one hour at 100 V and stained with ethidium bromide. The DNA bands in the gel were observed under UV transilluminator filter. The bands were photographed using a digital camera. Solis BioDyne 100 bp DNA Ladder (100-3000 bp) was used as a size markers.

Data analysis

Amplified fragments were considered as a binary character for the present (scored 1) and absent (Scored 0) using Total lab TL 120. Similarities were estimated by Jaccard's coefficient. Cluster analysis was carried out with NTSYS-pc software, UPGMA algorithm (Rohlf, 2000).

RESULTS

Variance estimation and mean performance

Combined analysis for seed yield ha⁻¹ across environments showed significant ($p < 0.05$) difference for genotypes and environments and genotype by environment interaction ($G \times Y$). The data in Table 3 revealed that the mean performance of the lines C1.5, C3.8, C6.3, and C1.6 for seed yield ha⁻¹ was higher than C by 3.4, 2.8, 0.5 and 16.7%. Seed yield ha⁻¹, on average, varied from 947.3 kg (C) to 1105.2 kg (C1.6). A high seed yield ha⁻¹ across environments was obtained by C1.6 compared with overall other genotypes. The genotypes ranked based on seed yield ha⁻¹ across environments to determine which genotypes most affected by environmental conditions (Table 4). Line C1.6 achieved the highest rank and achieved the lowest value of SD of the ranks compared to other genotypes.

Table 3. Combined analysis and mean performance of genotypes sesame lines for seed yield ha⁻¹.

| S.V. | DF | Mse | A | | B | | | | \bar{x} |
|-------|----|------------|------|----------------|----------------|----------------|-----------------|--------|-----------|
| | | | G | F ₈ | F ₈ | F ₉ | F ₁₀ | | |
| E | 3 | 135474.6** | C | 1505.6 | 552.0 | 853.3 | 878.4 | 947.3 | |
| G | 4 | 87478.2** | C1.5 | 1784.8 | 892.4 | 593.4 | 646.8 | 979.3 | |
| G * E | 12 | 86742.2** | C3.8 | 1449.0 | 558.9 | 1078.7 | 808.5 | 973.8 | |
| | | | C6.3 | 1046.5 | 632.5 | 1064.9 | 1063.8 | 951.9 | |
| | | | C1.6 | 1851.5 | 614.1 | 920.0 | 1035.0 | 1105.2 | |
| | | | SD | 319.8 | 139.9 | 197.3 | 171.1 | - | |
| | | | CV% | 8.0 | 17.0 | 19.0 | 17.0 | - | |

S.V.: source of variance, DF: degree of freedom, Mse: mean square, G: genotypes, A: AL-Sharqia, B: AL-Nubria, F: filial generation, \bar{x} : mean performance of lines, SD: standard deviation, CV%: coefficient of variability, ***P* < 0.05

Table 4. The standard deviation of rank (SD) of genotypes for seed yield ha⁻¹ in three generations.

| Genotypes | Generations | | | | SD |
|-----------|----------------|----------------|----------------|-----------------|------|
| | F ₈ | F ₈ | F ₉ | F ₁₀ | |
| | A | B | | | |
| C | 3 | 5 | 4 | 3 | 0.96 |
| C1.5 | 2 | 1 | 5 | 5 | 2.06 |
| C3.8 | 4 | 4 | 1 | 4 | 1.50 |
| C6.3 | 5 | 2 | 2 | 1 | 1.73 |
| C1.6 | 1 | 3 | 3 | 2 | 0.95 |

A: AL-Sharqia, B: AL-Nubria, SD: standard deviation

Molecular analysis

RAPD and ISSR techniques were used to detect markers linked to *Fusarium* wilt resistance and determine the genetic relationships among the genotypes under study. The lines were classified as highly resistant to *Fusarium* wilt at seedling and mature stages except line C1.5 (Shabana et al., 2014). So, according to their resistance, the C3.8 line was chosen as the highest

resistant one, while C was chosen as the susceptible one to *Fusarium* wilt.

RAPD analysis

Of the twenty RAPD primers used seven primers revealed a polymorphism, which only three developed markers linked to *Fusarium* wilt resistance as shown in Fig.1 and summarized in Table 5.

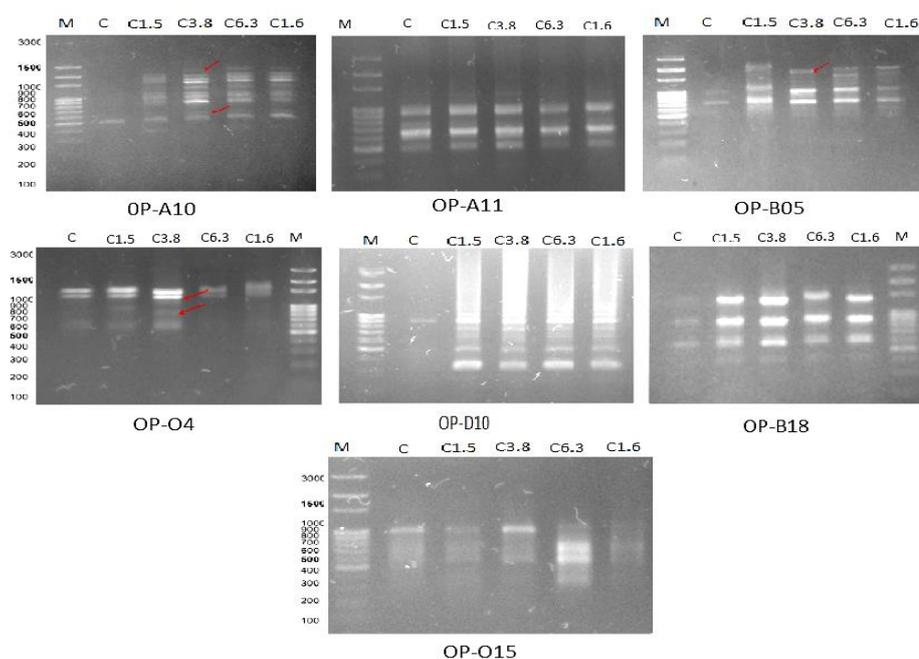


Fig. 1. Banding patterns of RAPD fragments of the five tested sesame (M= Marker, C, C1.5, C3.8, C6.3 and C1.6).

Table 5. RAPD markers and its amplification results.

| Primer name | TB | PB | P% | MS | MT | Size range | Primer |
|--------------|----|----|-------|-------------|--------|------------|--------|
| OP-A10 | 9 | 8 | 88.90 | 2672 652 | P P | 571-2672 | OP-A10 |
| OP-A11 | 5 | 2 | 40.00 | - | - | 402-1268 | OP-A11 |
| OP-B05 | 5 | 2 | 60.00 | 2036 | P | 767-2140 | OP-B05 |
| OP-B18 | 5 | 2 | 40.00 | - | - | 454-1723 | OP-B18 |
| OP-D10 | 6 | 5 | 83.33 | - | - | 311-1217 | OP-D10 |
| OP-O04 | 7 | 2 | 85.71 | 1231 865 | P P | 600-1634 | OP-O04 |
| OP-O15 | 4 | 0 | 25.00 | - | - | 379-1041 | OP-O15 |
| Total | 41 | 21 | | | 5 | | |

TB: total number of bands, PB: polymorphic bands, %P: % polymorphism, MS=molecular size, MT=marker type, P= Positive marker, N= negative marker.

OP-A10, OP-B05, and OP-O04 primers exhibited five positive markers with molecular sizes of 2672 bp, 652bp for OP-A10, 2036 bp for OP-B05 and 1231 bp, 865 bp for OP-O04 which were found in the line C3.8, while they were absent in C. RAPD markers were revealed 41 bands of which 21 (51.2%) polymorphic bands. The number of bands varied from 4 (for the OP-O15) to 9 (for the OP-A10 primers), ranging in size from 311 to 2672bp. The polymorphism percentages were ranged from 25% (for the OP-O15) to 88.9% (for the OP-A10) with an average of 60.42%.

The genetic similarity coefficient varied from 0.58 between C3.8 and C to 0.92 between C1.6 and C6.3 (Table 6).

Table 6. The genetic similarity matrix of the five samples based on RAPD markers.

| Genotypes | C | C1.5 | C3.8 | C6.3 | C1.6 |
|-----------|------|-------|------|------|------|
| C | 1 | | | | |
| C1.5 | 0.66 | 1 | | | |
| C3.8 | 0.58 | 0.865 | 1 | | |
| C6.3 | 0.60 | 0.84 | 0.89 | 1 | |
| C1.6 | 0.61 | 0.82 | 0.87 | 0.92 | 1 |

The constructed dendrogram obtained by UPGMA analysis divided these samples into two main clusters (Fig. 2).

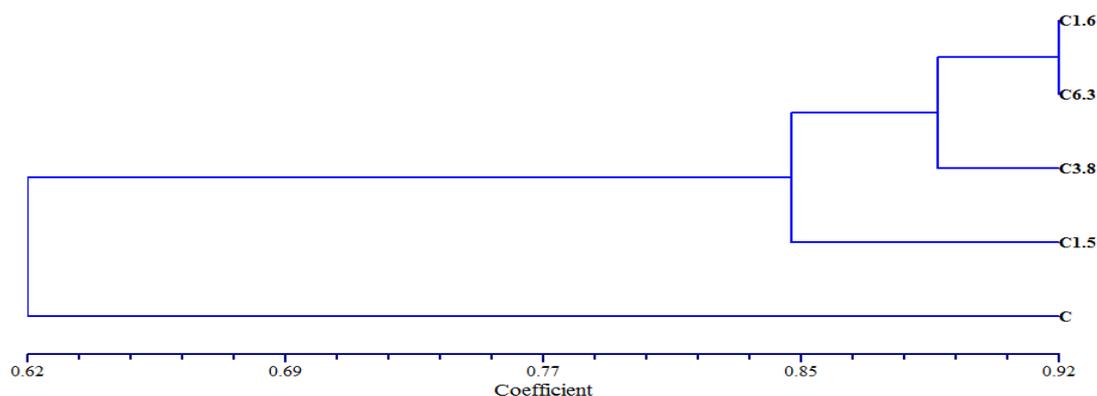


Fig. 2. UPGMA dendrograms of the five samples based on the genetic similarity matrix obtained with the Jaccard's index for the data from RAPD markers.

The first one included only C. The other cluster was divided into two main sub-clusters; the first one included only C1.5, while the other one was divided into two sub-sub clusters, the first one included only C3.8, while the second one included C1.6 and C6.3. According to RAPD analysis, these results indicated that the most closely related samples C1.6 and C6.3, which were located in the

same sub-sub cluster, while the most dissimilar samples were C3.8 and C.

ISSR analysis

The five ISSR primers revealed a polymorphism, only two primers developed markers linked to *Fusarium* wilt resistance as shown in Fig. 3 and summarized in Table 7.

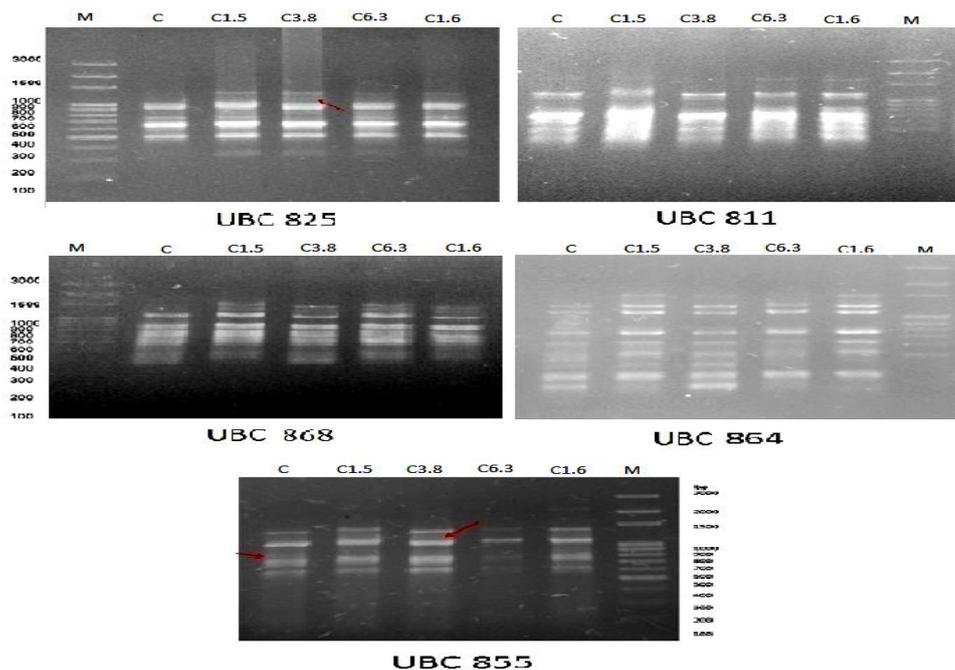


Fig. 3. Banding patterns of ISSR fragments of the five tested sesame (M= Marker, C, C1.5, C3.8, C6.3 and C1.6).

Table 7. ISSR markers and its amplification results.

| Primer name | TB | PB | P% | MS | MT | Size range |
|--------------|-----------|-----------|-------|------|----------|------------|
| UBC-811 | 10 | 4 | 50.00 | - | - | 483-1715 |
| UBC-825 | 10 | 3 | 40.00 | 1159 | P | 311-1284 |
| UBC-855 | 8 | 1 | 62.50 | 1242 | P | 574-2121 |
| UBC-864 | 9 | 3 | 33.33 | - | - | 293-1593 |
| UBC-868 | 8 | 4 | 50.00 | - | - | 371-1301 |
| Total | 45 | 15 | | | 3 | |

TB: total number of bands, PB: polymorphic bands, %P: % polymorphism, MS:molecular size, MT:marker type P: Positive marker, N= negative marker.

UBC-825 primer exhibited one positive marker linked to resistance with molecular sizes of 1159bp as well as UBC-855 primer exhibited one positive marker with molecular sizes of 1242bp linked to resistance were found in line C3.8 and one negative marker linked to C with molecular sizes of 801bp. A total of 45 bands were produced of which 15 (33.33%) polymorphic bands. The number of ISSR bands varied from 8 (for two primers UBC-855 and UBC-868) to 10 (for two primers UBC-811 and UBC-825), ranging in size from 293 to 2121 bp. The polymorphism percentages were ranged from 33.33% (for the UBC-864) to 62.50% (for the UBC-855) with an average of 47.17%.

The genetic similarity coefficient varied from 0.78 between C and C1.5 to 0.95 between C1.6 and C6.3 (Table 8).

Table 8. The genetic similarity matrix of the five samples based on ISSR

| Genotypes | C | C1.5 | C3.8 | C6.3 | C1.6 |
|-----------|------|------|------|------|------|
| C | 1 | | | | |
| C1.5 | 0.78 | 1 | | | |
| C3.8 | 0.87 | 0.87 | 1 | | |
| C6.3 | 0.85 | 0.85 | 0.90 | 1 | |
| C1.6 | 0.81 | 0.87 | 0.89 | 0.95 | 1 |

The dendrogram constructed from ISSR analysis (Fig. 4) was similar to the RAPD dendrogram. These results indicated that the most closely related samples were C1.6 and C6.3, while the most dissimilar samples were C and C1.5 which located in the two different main clusters.

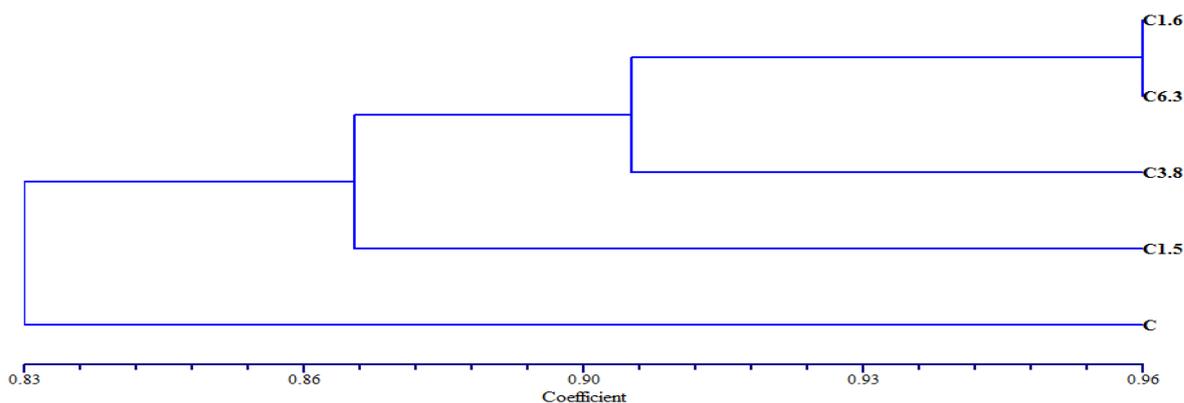


Fig. 4. UPGMA dendrograms of the five samples based on the genetic similarity matrix obtained with the Jaccard's index for the data from ISSR markers.

Combination of RAPD + ISSR analysis

The genetic similarity coefficient varied from 0.72 between C and C1.6 to 0.94 between C1.6 and C6.3 (Table 9). The RAPD and ISSR data were also combined for UPGMA cluster analysis (Fig. 5). The dendrogram was similar to the RAPD and ISSR dendrograms. According to these results, the most closely related samples were C1.6 and C6.3, while C and C1.6 were the most genetically distant.

Table 9. The genetic similarity matrix of the five samples based on RAPD + ISSR markers.

| Genotypes | C | C1.5 | C3.8 | C6.3 | C1.6 |
|-----------|------|------|------|------|------|
| C | 1 | | | | |
| C1.5 | 0.73 | 1 | | | |
| C3.8 | 0.75 | 0.86 | 1 | | |
| C6.3 | 0.74 | 0.85 | 0.90 | 1 | |
| C1.6 | 0.72 | 0.85 | 0.88 | 0.94 | 1 |

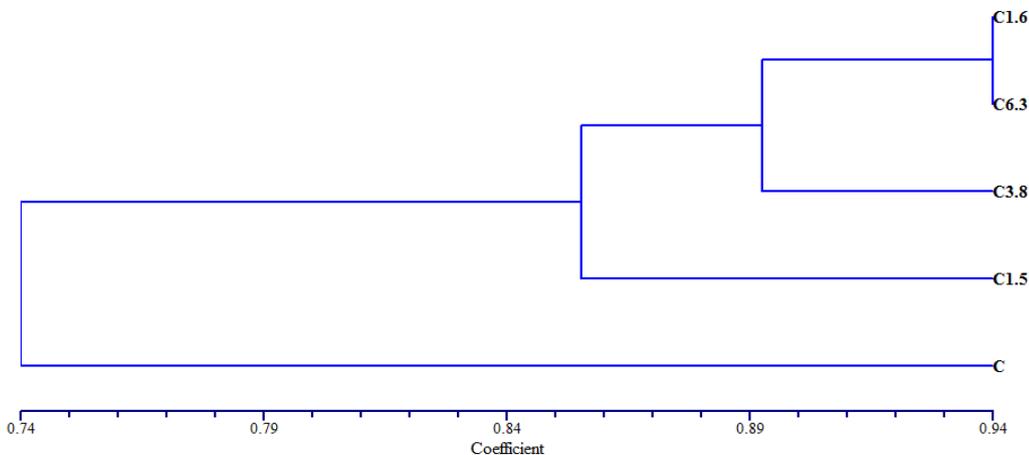


Fig. 5 UPGMA dendrograms of the five samples based on the genetic similarity matrix obtained with the Jaccard's index for the data from RAPD+ISSR markers.

DISCUSSION

Variance and Mean performance

One of the effective ways to keep crop production under new challenges of climate change is through varieties developed by plant breeding. The high-yield and disease-resistant varieties are the most important achievements of plant breeding, which adds more arable land to world agricultural production. The breeding process consists of three phases: creates variation (one year), line fixation (F_2 to F_6 generations), and field trials (Holbrook and Culbreath, 2007; Tillman and Gorbet, 2009; Newton et al., 2011). In this study, we have reached

the phase of field trials of advanced breeding lines (completely homozygous) which usually requires two years of testing against the commercial variety. In addition, breeders have always used modern plant breeding with traditional plant breeding to make their goals quickly (Forster et al., 2014). The data in Table 3 illustrated that the response of the genotypes is irresolute and fluctuated in seed yield ha^{-1} across environments through the growing seasons confirmed by genotype x environment interaction ($G \times Y$). And the differences were due to both of the main and interaction effects caused different fertility of soils (clay and sandy) irrigation methods (surface and sprinkler) and differences in the

genetic make-up of the genotypes (John et al., 2001; Hoballah et al., 2009; Ahmed and Ahmed, 2012; Baraki and Berhe, 2019). Environments represent the largest proportion of sums of squares followed by genotypes consequently, the possibility of developing new varieties for specific adaptation to each of these environments (Kebede et al., 2017; Mansour et al., 2018). The mean performance of lines for seed yield ha⁻¹ in AL-Sharqia was higher than AL-Nubria, because that the clay soil is higher fertility than sandy soil proven by a high percentage of organic matter in clay soil than sandy soil (Table 2). Two lines C1.6 and C1.5 excelled in the first season in clay soil while in sandy soil line C1.5 excelled. The line C1.6 achieved the highest seed yield ha⁻¹, on average, while the other lines and C were about equal. This line would have excellent gene combinations, be adapted to different environments. And many authors (Taghouti et al., 2017; Anter and Ashraf, 2018; Baraki et al., 2019) pointed out to that sesame genotypes were differences in seed yield across by changing in environments. Moreover, the result of rank for genotypes based on seed yield ha⁻¹ across environments in Table 4 showed that C and C1.6 were less affected by environmental conditions indicated by high rank and lower SD values (Karimizadeh et al., 2012). Apparently, line C1.6 showed a high response for seed yield ha⁻¹ across environments and could be classified as a productive and stable genotype. This was probably due to its ability to produce secondary metabolites and stored carbon as a strategy to respond to different environments (Bennett and Wallsgrove, 1994; Harborne, 1999; Beckman, 2000; Kroymann, 2011; Li et al., 2018) so, could be used as a parent in the other breeding program to form new varieties that more fixability for tackling climatic change.

Molecular analysis

The molecular markers could be utilized to enhance conventional plant breeding strategies of disease resistance. Therefore, this study carried out for the selection of the resistant and susceptible lines to *Fusarium* wilt through a set of RAPD and ISSR markers to detect marker linked to *Fusarium* wilt resistance to enhance resistant germplasm resources for increasing yield of sesame. The extent of polymorphism in this study was ranged from 25.00 to 88.90% in the case of RAPD was greater than in of the ISSR (33.33–62.50%). These differences in polymorphism may be due to the differences in the amount of genetic variation between the samples under the study as indicated by Poerba and Ahmad (2010). Thus, this study indicates that samples examined have high genetic variation. These results complied with Ercan et al. (2004) showed that RAPD markers revealed polymorphism level of 78% was reported in 38 accessing of sesame in Turkey and 73% polymorphism was reported in a core collection of sesame from China by Zhang et al. (2010). RAPD markers revealed more polymorphism than ISSR markers in different plant species (Farajpour et al., 2011; Patel et al., 2016). Our results showed that the average of

polymorphism revealed in ISSR markers was 47.17% was higher compared with Kim et al. (2002) in which the level of polymorphism among 75 Korean sesame genotypes was 33.0% with 14 ISSR markers. However, our results were low as compared with the 98.5% of Anitha et al. (2010) in 10 sesame varieties of Tamil Nadu. And 76.47% of Parsaeian et al. (2011) in a study of genetic variation in sesame with 13 ISSR primers. Admas et al. (2013) detected that the polymorphism was 97.3% between six sesame genotypes. Also, Kurt and Arioglu (2018) detected polymorphism percentage ranged from 33% to 100% with an average of 68.2%. This difference between our results and the previous studies may be due to the low numbers of primers used. Whilst, the use of a few primers will be sufficient when the variation between genotypes is high (Li and Midmore, 1999). Salahlou et al. (2019) have been used the same number of ISSR primers to detect genetic diversity among *Macrophomina phaseolina* isolates of sesame in Iran. From the previous results, it was clear that the dendrogram constructed using RAPD and ISSR markers were highly similar. The genetic similarity coefficient observed in this study, 0.58 - 0.92 in the case of RAPD; 0.78 - 0.95 in the case of ISSR and 0.72- 0.94 in case of pooled data of RAPD and ISSR. These results are confirmed by Dar et al. (2017) reported that the similarity coefficient of RAPD ranged from 0.51 to 0.88, and ranged from 0.34 to 0.89 between 15 sesame accessions by Quenum and Yan (2017). Whilst, our results were high compared with previous studies by Parsaeian et al. (2011) in which the coefficient of similarities ranged from 0.09 to 0.55. In this study, we found five positive RAPD markers (OP-A10_{2672& 652}; OP-B05₂₀₃₆, and OP-O04_{1231& 865}) and two positive ISSR markers (UBC-825₁₁₅₉ and UBC-855₁₂₄₂) and one negative marker (UBC-855₈₀₁) linked to *Fusarium* wilt resistance. The obtained results were in a good line with those obtained by Singh et al. (2011) used RAPD molecular markers in finding markers linked to *Fusarium* wilt resistance gene in castor bean and found three RAPD markers (RKC 231375, RKC 211080 and OPBE 18900) linked to the resistance genes. Haji-Allahverdipoor et al. (2011) found three ISSR markers (864₄₀₀, UBC-811₁₂₅₀ and UBC-811₆₅₀) in chickpea. And Maisuria et al. (2017) found three markers (CS-27₇₀₀, UBC-825₁₂₀₀, and UBC-170₅₀₀) in chickpea. In general, the results indicated that the four lines of sesame, on average, surpassed C in seed yield ha⁻¹. Besides, these lines characterized by high resistance to *Fusarium* wilt except line C1.5 (Shabana et al., 2014).

CONCLUSIONS

The study concluded, from the results into that sesame genotypes significantly differ (P<0.05) across environments, where the differences were due to both of the main and interaction effects and environments contribute to a high proportion of variation, followed by genotypes, and thus the ability to develop new varieties that adapt to each of these environments. Line C1.6 achieved the highest seed yield ha⁻¹, on average, compared

to check variety also, it achieved less value of the standard deviation of ranks through environments. According to molecular marker analysis. RAPD and ISSR markers revealed seven positive markers linked to *Fusarium* wilt resistance found in C3.8. And, one negative marker linked to *Fusarium* wilt resistance found in C therefore, which could be considered as reliable markers for *Fusarium* wilt resistance in sesame. Therefore, it is possible to combine the high- yielding and *Fusarium* wilt resistance in sesame.

ACKNOWLEDGMENT

Many thanks to the National Research Center, Egypt for funding this study.

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