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## Germination Characters as Affected by Seed Priming of some Turnip and Radish Native Cultivars under Different Levels of Salinity

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### ABSTRACT

A laboratory experiment was conducted at vegetable seed production laboratory in Sabahiya Horticulture Research station during February until August 2021, to study induce salinity tolerance in some native cultivars of turnip and radish through seed priming with solution of ( $H_2O_2$ ,  $KNO_3$ , NaCl and tap water), then examine the possibility of seed priming on the germinate under different levels of NaCl. Finally study the genetic diversity of turnip and radish cultivars and genetic relationships among the genotypes based on molecular characterization using RAPD - PCR analysis. Results showed varietal differences on germination parameters among the five-turnip and radish genotypes. In general, germination parameters were decreased as salt level increased up to 4000 ppm NaCl, however, control treatment was significantly equal to that obtained by salt level at 2000 NaCl in some germination parameters, Effect of seed priming substances on germination parameters were differed, generally, seed priming with  $H_2O_2$  (0.5%) or tap water gave the best results, meanwhile, seed priming with NaCl (3%) gave the worst results. Regarding the study of genetic diversity among the studied Brassicaceae family members, a cluster analysis divided the five turnip genotypes into two main groups and one sub-cluster. Concerning the five radish genotypes, cluster analysis divided the genotypes into two main groups with one sub- cluster. It can be concluded that the morphological and molecular markers could be a better tool for studying the genetic diversity.

**Keywords:** turnip, radish, seed priming, salt level, RAPD

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### 1. Introduction

Egypt had been gifted a tremendous treasure of Flora and Fauna, including some varieties of vegetables. However, it subjects to either extinction or stolen by other countries. These varieties could had special characteristics such as tolerance to biotic and abiotic stress. Thus, this causes great loss for national economy. In this respect, most of native cultivars of turnip and radish varieties are not registered (named balady).

Turnip (*Brassica rapa* L.,  $2n=20$ ) and radish (*Raphanus sativus* L.,  $2n = 18$ ) are belong to Brassicaceae family, they rich in vitamins, minerals (such as calcium, potassium, iron, copper, magnesium and zinc) and anti-oxidants (Jeong *et al.* 2016).

Radish belongs to a genus different from that of turnip, but they are highly similar in morphology to each other as vegetables. Shapes of seed and sizes are obviously different between them. Although the draft genome sequences of Chinese cabbage in *B. rapa* have been obtained and published, it is difficult to use these sequence data as references to determine the radish genome sequences because of highly complicated genome synteny between *B. rapa* and *R. sativus* (Li *et al.*, 2011)

Abiotic stresses such as salt and drought are among factors limiting plant productivity (Bohnert *et al.*, 1999). High salinity in soil or irrigation water is a common environmental problem affecting seed germination and plant growth. Soil salinity may affect the germination of seeds either by creating an osmotic potential external to the seed preventing water uptake, or through the toxic effects of  $Na^+$  and  $Cl^-$  ions on the germinating seed (Farhoudi *et al.*, 2011). Salt and osmotic stresses are responsible for both inhibition or delayed seed germination and seedling establishment (Almansouri *et al.*, 2001).

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Seed priming is a pre-sowing treatment in different ways so as to cause early germination and obtain better seed vigor. Priming improves seed viability, synchronizes and accelerates germination and sprouting, increases stress resistance and antioxidant activity, and improves plant productivity and growth (McDonald, 2000). Specifically, under stress conditions, it induces the germination changes which are maintain the germination rate and uniformity in the seedling emergence (Ashraf and Fooland, 2005).

Seed priming with H<sub>2</sub>O<sub>2</sub> (halopriming) having the capacity to enhance the multi-resistance to heat, drought, chilling and salt stress. H<sub>2</sub>O<sub>2</sub> may plays an important role in signal transduction for abiotic stress tolerance, although H<sub>2</sub>O<sub>2</sub> is toxic at high concentrations (Uchida *et al.*, 2002). To improve the stress tolerance in plants, controlling the free radicals is the better way (Bor *et al.*, 2003). Increasing production of hydrogen peroxide leads to improve the survival of the crop plants.

Seed priming with NaCl and KNO<sub>3</sub> (Osmo-priming) is the most common method of priming used by solutions with low osmotic potentials. In this method many compounds have been used to achieve a solution or known water potential (Farooq *et al.*, 2006). In primed seeds usually high germination rate, high germination uniformity and in some cases high final germination was shown (Basra *et al.*, 2005).

Tap water (hydro-priming) is the most practical technique without much labor cost and disposal concern associated with other priming agents. Hence, the amount of water that initiates the metabolic events to a point short of radical emergence, the method of water absorption, and duration of its absorption are important considerations for seed quality enhancement and synchronization of the germination process through hydro-priming (Srinieng *et al.*, 2015).

Random amplified polymorphic DNA (RAPD) markers are widely used in genetic studies due to their simplicity and speed. RAPD analysis is one of the most straight forward marker-based technique among all the accessible marker techniques which are executed to evaluate the composition, modifications, and advancement of genetic material (Raza A2020).

Present study was conducted to induce salinity tolerant in some native cultivars, of turnip and radish through seed priming with solution of (H<sub>2</sub>O<sub>2</sub>, KNO<sub>3</sub>, NaCl and tap water). Then examine the possibility of seed priming on germinate under different level of NaCl. Finally study the genetic diversity of turnip and radish cultivars and genetic relationships among the genotypes sampled based on molecular characterization using RAPD - PCR analysis.

## **2. Materials and Methods**

A laboratory experiment was conducted at vegetable seed production laboratory in Sabahiya Horticulture Research station from February until August 2021, to study the response of seed priming of some native cultivars of turnip and radish to germinate under different salinity concentrations and study genetics identification using RAPD - PCR analysis.

### **2.1. Plant material and experimental design:**

Mature seeds of five Egyptian native cultivars (named balady) of both radish and turnip collected from Alexandria, Kafr El-Sheikh, ELwady Elgadid, Gharbiya and Assiut governorates. The experiment was laid out in factorial completely randomized design with three replications. The experiment includes three factors. The first one includes five genotypes of both radish and turnip. The second factor includes priming in (3%NaCl, 3%KNO<sub>3</sub>, 0.5% H<sub>2</sub>O<sub>2</sub> and tap water in addition to non-priming seed). The third factor included two different NaCl levels (2000, 4000 ppm) plus the control (tap water). Thus, the total number of treatments were 75 treatments (5 cultivars×5 seed priming×3 NaCl levels) for both turnip and radish. The collected data were analyzed using analysis of variance (ANOVA) techniques using F test. The differences between the treatment means were determined by using Duncan's New Multiple Range at 5% level of significance (Snedecor and Cochran, 1982).

### **2.2. Seed priming procedures:**

Seeds of both variety were primed with (3%NaCl, 3%KNO<sub>3</sub>, and 0.5% H<sub>2</sub>O<sub>2</sub>) solutions for 6 h. for hydro-priming seeds were soaked in tap water for the same duration. Some seeds were untreated (control) the prime seeds were dried overnight.

**2.3. Seed germination under salt treatments:**

The germination test was conducted using Petri dish as substrate. Hundred seeds for both variety were placed in each Petri dish. Tap water was used as control. The Petri dish were observed every day and respective solutions were supplied whenever required for 7 days.

**2.4. Data collection**

Germination parameters: are calculated as described in the Association of Official Seed Analysts (AOSA, 1983 and 1991).

$$\text{Germination (\%)} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds placed in petridish}} \times 100 \dots\dots\dots(1)$$

$$\text{Germination speed (\%)} = \frac{\text{Number of germinated in 24h}}{\text{Number of germinated in 120h}} \times 100 \dots\dots\dots(2)$$

$$\text{Germination index (GI)} = G1/1+G2/2+\dots +Gi/i; \dots\dots\dots(3)$$

where G1 is number of germinated seeds at day 1, G2 is number of germinated seeds at day 2; and so on.

Seven days later after experiment started, the following parameters were measured:

Seedling length (cm): Randomly selected five seedlings were taken from each Petri dish to measure seedling length (stem and root).

Seedling dry matter%: Two grams of each genotype's seedling were weighted and let till dry in oven at 45° C for one day then the percentage of dry matter was taken.

**2.5. PCR based on RAPD Analysis:**

Genomic DNA isolation: Genomic DNA was extracted from the young leaves of the five turnip and radish genotypes by employing genomic DNA extraction kit (Easy Pure Plant Genomic DNA Kit) DNA samples were stored at -20°C. DNA quality was checked by electrophoresis in a mini gel.

In the present study, two different RAPD primers were employed to evaluate the efficiency of these markers in diversity analysis of turnip and radish genotypes. The sequences of the used primers are shown in Table 1. PCR reactions were performed in 20µl total volume, using 1µl of diluted DNA, 1µl of each primer for the amplification reaction, 10µl master mix (Taq Ready Mix PCR Kit from the fast gene), and 8µl ddH<sub>2</sub>O (sterile water) for all reactions. The tubes were capped and placed in a thermocycler and the cycling was started immediately. Amplification protocol was carried out using PCR cycler 600 programmed for initial denaturation step at 94°C for five min, followed by 40 cycles each at 94°C for 30 sec, annealing at 37°C, and extension at 72°C for 1min.

**Table 1:** Code and sequences of the 2 different RAPD primers used in the present study

Primers	Sequence (5'-3')
OPA2	GTG ATC GCAG
OPA07	GAAAGGGGTG

The products of RAPD - based PCR analyses were detected using agarose gel electrophoresis (1.5% in 1X TBE buffer) stained with ethidium bromide (0.3µl). PCR products were visualized on U.V. light; photographed and analyzed using the Total Lab Quant software program.

For molecular data and cluster analysis, data were scored for computer analysis on the basis of the presence of the amplified products for each primer. If a product was present in a genotype, it was designated as “1”, if absent, it was designated as “0”, after excluding the unreproducible bands. Pair-wise comparisons of genotype, based on the presence or absence of unique and shared polymorphic products, were used to determine similarity coefficients, according to (Jaccard, 1908). DNA fragment size was estimated by comparison with a 1-kbp DNA ladder (Ready to use from Gene Direx). The similarity coefficients were then used to construct dendograms, using the Unweighted Pair Group Method with Arithmetic Averages (UPGMA) employing the SAHN (Sequential, Agglomerative,

Hierarchical, and Nested clustering) from Past program version 4.03. Comparing between morphological and molecular markers were studied.

### 3. Results and Discussion

#### 3.1. Germination parameters

##### 3.1.1. Turnip

##### 3.1.1.1. Main effect

Results in Table (2) show varietal differences on germination parameters among the five-turnip genotypes. Gharbiya and Elwady Elgadid genotypes gave the highest percentage of germination. Meanwhile, the genotype of Assiut had the lowest mean value of germination speed (83.0) compared with the other genotypes. Elwady Elgadid genotype gave the highest mean value of germination index and seedling dry matter percentage. Regarding seedling length, there was no significant differences among all the genotypes except that of the Gharbiya genotype which gave the shortest seedling. The differences between genotypes in germination might be due to genetically factors and heredity (Ghazizade *et al.*, 2012).

**Table 2:** Varietal differences on germination parameters among the five turnip genotypes overall seed priming and salt levels.

Genotypes	Germination %	Germination speed %	Germination index	Seedling length (cm)	Seedling dry matter %
Alexandria	84.0c <sup>#</sup>	77.4d	77.6d	4.5ab	5.3b
Kafr El-Sheikh	86.3b	79.8c	81.2b	4.7a	4.8d
Elwady Elgadid	87.7a	81.7b	82.8a	4.1ab	5.9a
Gharbiya	88.2a	79.0cd	81.6b	3.8b	4.8d
Assiut	83.0d	84.4a	80.0c	4.2ab	4.9c

<sup>#</sup>Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

The effect of salt levels on germination parameters is illustrated in Table (3). Generally, all germination parameters as decreased as the salt level increased up to 4000 ppm NaCl. However, there were no significant differences between control treatment (tap water) and salt level at 2000 ppm NaCl concerning germination speed and seedling length. According to Hakim *et al.* (2010), any crops growing under salinity conditions there may be a reduction in seedling length. Plants display great diversity concerning salinity tolerance. Species distribution and survival mainly depend on the seed's ability to complete germination and the seedling's ability to develop successfully under unfavorable conditions (Živković *et al.*, 2007).

**Table 3:** Effect of salt levels on germination parameters overall turnip genotypes and seed priming.

Salt level (ppm)	Germination %	Germination speed%	Germination index	Seedling length (cm)	Seedling dry matter %
2000 ppm	86.9 b <sup>#</sup>	82.5a	80.4b	4.6a	5.3b
4000 ppm	80.6c	75.4b	77.2c	3.5b	3.9c
Control	90.0a	83.4a	84.0a	4.7a	6.3a

<sup>#</sup>Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

Effects of seed priming substances on germination parameters are shown in Table (4). The highest percentage of germination was obtained by seed priming with tap water and there aren't significant differences between H<sub>2</sub>O<sub>2</sub> at concentration 0.5% and tap water treatment (98.4 and 98.6 %, respectively). Seed priming with H<sub>2</sub>O<sub>2</sub> at concentration 0.5% gave the highest germination speed (97) and there are no significant differences between H<sub>2</sub>O<sub>2</sub> and KNO<sub>3</sub> treatments. On the other hand, Seed priming with NaCl at concentration 3% had the lowest mean values for all germination parameters comparing with the other treatments. Control (without treatment) gave the longest seedling (5.1 cm), comparing with the other treatments. Tap water treatment gave the highest dry matter percentage and germination index. Priming techniques are being used to improve seed germination under both optimal

and adverse conditions (Jisha *et al.*, 2013). The positive effects of priming could even be clearer under unfavorable than favorable conditions (Chen and Arora, 2013).

**Table 4:** Effect of seed priming substances on germination parameters overall turnip genotypes and salt levels.

Seed priming substances	Germination %	Germination speed%	Germination index	Seedling length (cm)	Seedling dry matter %
H <sub>2</sub> O <sub>2</sub> (0.5%)	98.4ab <sup>#</sup>	97.0a	95.2b	4.3b	5.9b
KNO <sub>3</sub> (3 %)	96.8c	96.3ab	94.0c	4.1b	4.8c
NaCl (3 %)	37.7d	20.7d	21.2d	3.4c	4.1d
Tap water	98.6a	93.5c	96.4a	4.5b	6.2a
Without	97.0b	94.7bc	96.0a	5.1a	4.8c

<sup>#</sup>Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

### 3.1.1.2. Interaction effects

Results in Table (5) exhibit the effect of interaction between the five turnip native genotypes and salt levels on germination parameters overall seed priming. The interaction was significant or highly significant for all germination parameters. Balady Gharbiya gave the highest percentage of germination by the salt level at 2000 ppm treatment (93.1%) and Control (tap water, 93.7%). The highest mean value of germination index was obtained by Control (tap water) for balady Gharbiya (87.6). Meanwhile, the most effective germination speed was obtained for balady Assiut by tap water (control, 87.50) followed by the salt level at 2000 ppm treatment (86.4). The highest seedling length mean value was obtained by control (tap water) treatment for balady Assiut compared with the other genotypes affected by salt levels. Balady of Elwady Elgadid gave the highest dry matter percentage by control treatment. Although salinity stress mostly reduces the germination percentage and delays the onset of germination. Its effects are modified by interactions with other environmental factors as temperature and light. Salinity can affect germination by affecting the osmotic component, which is that the ionic component, i.e., Na and Cl accumulation (Živković *et al.*, 2007).

**Table 5:** Effect of interaction between turnip genotypes and salt levels on germination parameters overall seed priming.

Genotypes	Germination %**			Germination index*			Germination speed%**		
	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control
	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)
Alexandria	82.5f	78.4hi	91.2b	74.8g	74.8g	82.8bcde	79.7ef	71.5h	80.9def
Kafr El-Sheikh	89.3c	81.7fg	87.9d	82.4cde	78.4f	82.4cde	83.4bcd	75.7g	80.4def
Elwady Elgadid	88.3cd	85.9e	88.9cd	82.8bcde	82.0e	84.0b	82.8def	79.9ef	82.5def
Gharbiya	93.1a	77.9i	93.7a	83.6bc	73.2h	87.6a	80.3def	70.7h	85.9abc
Assiut	81.4g	79.1hi	88.4cd	78.8f	78.4f	83.2bcd	86.4ab	79.2f	87.5a

Genotypes	Seedling length (cm)**			Seedling dry matter %**		
	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control
	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)
Alexandria	4.8def	4.0h	4.7efg	5.1e	4.5g	6.3c
Kafr El-Sheikh	5.1bc	4.0efg	5.2efg	4.8f	3.7h	5.8d
Elwady Elgadid	4.2bcd	3.7h	4.6fg	6.5bc	4.5g	6.9a
Gharbiya	4.1ab	3.0efg	4.4cde	4.9f	3.7h	5.8d
Assiut	4.6efg	3.1gh	5.0a	5.1e	3.2i	6.5b

\*,\*\*, Interaction of such parameter is significant at level 5% and 1% of probability, respectively.

<sup>#</sup>Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

The effect of interaction between seed priming substances and salt levels on germination parameters overall the five turnip genotypes are shown in Table (6). Interaction was highly significant for all germination parameters. These results indicated that seed priming substances ranked as different as salt levels concerning such parameters. Moghanibashi *et al.*, (2012) showed that primed seeds produced the higher germination index, germination rate days to 50% germination, and germination index than non-primed seeds under all salinity levels. Pahoja *et al.*, (2013) indicated that hydro-priming proved significantly better than osmo-priming (KNO<sub>3</sub>) under the wide range of salinity levels. The

germination percentage of a seed can be characterized by three parameters; time of germination, germination speed, and extent or capacity (cumulative germination percentage at the tip of the testing period). Germination parameters are useful for estimating the conversion of seeds to seedlings and, thus, the suitability of seed for commercial seedling production. Germination parameters are also useful in determining the type of seed pretreatment and incubation period needed to attain a high level of germination (Kolotelo *et al.*, 2001).

**Table 6:** Effect of interaction between seed priming substances and salt levels on germination parameters overall turnip genotypes.

Seed priming substances	Germination %**			Germination index**			Germination speed%**		
	2000 ppm NaCl	4000 ppm NaCl	Control (tap water)	2000 ppm NaCl	4000 ppm NaCl	Control (tap water)	2000 ppm NaCl	4000 ppm NaCl	Control (tap water)
H <sub>2</sub> O <sub>2</sub> (0.5%)	98.1bcd	97.5d	99.5a	90.3e	96.7c	98.1ab	98.3a	84.6c	97.7a
KNO <sub>3</sub> (3%)	97.7cd	95.2e	97.6d	96.1c	89.6e	96.8c	97.1a	88.7b	98.3a
NaCl (3%)	42.3g	15.9h	55.0f	23.8g	9.6h	30.3f	23.2d	12.7e	26.1d
Tab water	98.5abcd	98.3bcd	98.9abc	96.5c	96.2c	96.9bc	96.5a	95.8a	96.6a
Without	98.1bcd	96.2e	99.1ab	96.1c	94.2d	98.3a	97.4a	95.1a	98.4a
Seed priming substances	Seedling length (cm)**			Seedling dry matter %**					
	2000 ppm NaCl	4000 ppm NaCl	Control (tap water)	2000 ppm NaCl	4000 ppm NaCl	Control (tap water)			
H <sub>2</sub> O <sub>2</sub> (0.5%)	4.4bcd	3.9cd	4.6abcd	6.1e	4.5h	7.2b			
KNO <sub>3</sub> (3%)	4.4bcd	3.4d	4.5bcd	4.6h	3.4k	6.4d			
NaCl (3%)	4.2bcd	2.1e	4.0cd	4.1i	3.6j	4.4h			
Tab water	4.3bcd	4.0cd	5.1abc	6.8c	4.1i	7.8a			
Without	5.7a	4.3bcd	5.3ab	4.8g	4.1i	5.6f			

\*\* Interaction of such parameter is significant at level 1% of probability.

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

Concerning the effect of the interaction between seed priming treatments and studied genotypes results were illustrated in Table (7). Results revealed that final parameters were significantly affected with the interaction between cultivars and seed priming treatments. Concerning this respect, Kaya *et al.*, (2011) concluded that hydro priming increased germination and seedling growth under salt and drought stress. Bajehbaj (2010) reported that the germination percentage of primed seeds was greater than that of non-primed seeds.

### 3.1.2. Radish

#### 3.1.2.1. Main effect

Results in Table (8) show varietal differences in germination parameters among the five radish genotypes. Balady Gharbiya gave the highest mean values for all the studied germination parameters except for seedling dry matter percentage.

Meanwhile, balady Alexandria had the highest percentage of seedling dry matter compared with the other genotypes. The groups of plants that are well adapted to saline habitats are called halophytes. Their seeds germinate well in water and the germination is similar to that of seeds of non-adapted species. However, they differ from their inability to germinate at higher salt concentrations the soil. The salinity tolerance of the many perennial halophytes dependent on a variety of abiotic factors (Baskin and Baskin, 1998).

Effect of salt levels on germination parameters is illustrated in Table (9). Generally, all germination parameters decreased significantly as the salt level increased up to 4000 ppm NaCl. Plants overcome salinity-induced osmotic effects through the accumulation of inorganic or by the synthesis of organic solutes such as free proline, glycine betaine, and free amino acids by a process known to as an osmotic adjustment in response to the decreased external water potential (Zavariyan *et al.*, 2015).

**Table 7:** Effect of interaction between turnip genotypes and seed priming substances overall salt levels on germination parameters.

Seed priming substances	Germination %**					Germination index**					Germination speed%**				
	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut
<b>H<sub>2</sub>O<sub>2</sub> (0.5%)</b>	97.4def	99.5a	96.4fg	99.1abc	99.2ab	89.6f	96.7bc	94.6d	95.7d	98.0ab	85.2d	94.8ab	96.4ab	93.1bc	97.9a
<b>KNO<sub>3</sub> (3%)</b>	97.3ef	99.4a	99.0abcd	98.0abcdef	90.4h	95.4cd	97.6ab	98.4a	89.5f	89.4f	96.5ab	96.7ab	98.9a	83.3d	97.9a
<b>NaCl (3%)</b>	30.1i	37.4k	45.7j	48.8i	26.3m	15.5j	20.8i	25.8h	28.5g	15.3j	20.1fg	16.8g	17.0g	21.4f	28.0e
<b>Tab water</b>	97.7bcdef	98.2abcde	98.2abcde	99.3ab	99.3ab	92.2e	95.5cd	97.2ab	98.8a	98.8a	90.2c	95.1ab	98.1a	99.0a	99.1a
<b>Without</b>	97.5cdef	97.0efg	99.1abc	95.7g	99.5a	94.5d	94.5d	98.0ab	94.7d	98.8a	94.7ab	95.5ab	98.1a	97.8a	98.7a

Seed priming substances	Seedling length (cm)**					Seedling dry matter %**				
	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut
<b>H<sub>2</sub>O<sub>2</sub> (0.5%)</b>	4.4abc	4.4abc	4.2abc	4.0bcd	4.4abc	5.7e	6.4b	6.1c	5.7e	5.5f
<b>KNO<sub>3</sub> (3%)</b>	4.9abc	5.7a	4.8abc	4.7abc	5.2abc	5.9de	4.7hij	4.3ki	4.7h	4.5ijk
<b>NaCl (3%)</b>	4.0bcd	4.6abc	3.5cd	2.4d	2.4d	3.6n	3.7mn	4.4jk	3.8	4.6hi
<b>Tab water</b>	4.3abc	5.3ab	3.7bcd	3.9bcd	4.9abc	6.5b	5.2g	9.0a	4.5hijk	5.8de
<b>Without</b>	4.7abc	3.5cd	4.1abc	4.1abc	4.0bcd	4.6hij	3.9	6.0cd	5.3fg	4.2i

\*\* Interaction of such parameter is significant at level 1% of probability.

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability

**Table 8:** Varietal differences on germination parameters among radish genotypes overall seed priming and salt levels.

Genotypes	Germination %	Germination index	Germination speed%	Seedling length (cm)	Seedling dry matter %
Alexandria	90.7d#	86.0e	85.4d	3.8d	5.9a
Kafr El-Sheikh	93.2c	88.4d	86.5c	4.7c	5.6b
Elwady Elgadid	96.7b	90.8c	87.2c	5.2b	5.5c
Gharbiya	99.2a	96.8a	94.9a	5.9a	5.3d
Assiut	96.7b	93.2b	92.6b	5.1b	4.9e

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

**Table 9:** Effect of salt levels on germination parameters in radish genotypes overall genotypes and seed priming.

Salt level (ppm)	Germination %	Germination index	Germination speed%	Seedling length (cm)	Seedling dry matter %
2000 ppm	96.3b#	92.0b	89.7b	4.9b	5.3b
4000 ppm	92.0c	86.8c	85.2c	3.2c	4.6c
Control	97.6a	94.4a	93.1a	6.8a	6.4a

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

Effects of seed priming substances on germination parameters are shown in Table (10). Seed priming with H<sub>2</sub>O<sub>2</sub> gave the highest mean values for all studied parameters and there have been no significant differences between without treatment and H<sub>2</sub>O<sub>2</sub> treatment on germination percentage, and seedling length. Also, there have been no significant differences between tap water treatment and without treatment concerning the germination percentage and seedling length. Generally, these results indicated that all germination parameters were improved by priming seed especially H<sub>2</sub>O and H<sub>2</sub>O<sub>2</sub> (0.5%). El-Saidy *et al.* (2011) concluded that hydro priming increased germination and seedling growth under stress. Bajehbaj (2010) reported that the germination percentage and germination index of primed seeds was greater than that of non-primed seeds. Seed priming is one in every of the useful physiological approaches that would adapt species to saline conditions it's a simple, low cost and low-risk technique accustomed overcome the salinity problem in agricultural lands (Gholami *et al.*, 2015). EL- Saidy *et al.* (2011) reported that priming seed significantly improved germination percentage in cultivars.

**Table 10:** Effect of seed priming substances on germination overall radish genotypes and salt levels.

Seed priming substances	Germination %	Germination index	Germination speed%	Seedling length (cm)	Seedling dry matter %
H <sub>2</sub> O <sub>2</sub> (0.5%)	99.1a#	98.8a	99.0a	5.2a	6.1a
KNO <sub>3</sub> (3%)	98.0b	96.0c	95.9b	5.2a	5.1c
NaCl (3 %)	80.9c	64.4d	55.1c	3.8b	5.0c
Tap water	99.2a	99.2a	99.6a	5.2a	5.9b
Without	99.1a	97.2b	96.3b	5.3a	5.1c

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

### 3.1.2.2. Interaction effects

Results in Table (11) exhibit the interaction between the five radish genotypes and salt levels on germination parameters. Interaction was significant or highly significant for all germination parameters. These results indicated that radish genotypes ranked as different as salt levels. Plant species vary in how well they tolerate salt. Some plants will tolerate high levels of salinity while others can tolerate little or no salinity. The relative growth of plants in the presence of salinity is termed their salt tolerance. A high salt level interferes with the germination of seeds. Salinity acts like drought on plants, preventing roots from performing their osmotic activity, where water and nutrients move from an area of high concentration. Therefore, the salt levels in water cannot go in the plant roots (Bojović *et al.*, 2010).

**Table 11:** Effect of interaction between the five radish genotypes and salt levels overall seed priming.

Genotypes	Germination %**			Germination index**			Germination speed%**		
	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control
	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)
Alexandria	91.1g#	82.3i	98.6B	84.8i	79.2k	94.0e	81.4cd	84.5bcd	90.1abcd
Kafr El-Sheikh	94.7e	89.8h	95.0E	89.6h	85.6k	90.4g	87.3bcd	83.7bcd	88.5abcd
Elwady Elgadid	97.3c	95.9d	96.8C	92.0f	86.8i	93.6e	88.7abcd	80.1cd	92.8abc
Gharbiya	99.9a	98.3b	99.5A	97.6b	93.2e	99.6a	95.8ab	89.2abcd	99.8a
Assiut	98.4b	93.6f	98.0B	96.0i	88.8d	95.2c	95.4ab	88.3abcd	94.1ab
Genotypes	Seedling length (cm) **			Seedling dry matter %**					
	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control			
	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)			
Alexandria	3.65h	2.12j	5.86d	6.0de	4.5i	7.0a			
Kafr El-Sheikh	4.67e	2.83i	6.62c	4.7h	5.1f	6.1de			
Elwady Elgadid	5.17e	3.33h	7.17b	6.1de	3.9j	6.5b			
Gharbiya	5.93d	4.24g	7.53a	4.6hi	6.0e	6.3c			
Assiut	5.23e	3.33h	7.13b	4.5i	5.1f	6.1de			

\*\* Interaction of such parameter is significant at level 1% of probability.

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

Effect of interaction between seed priming substances and salt levels on germination parameters overall the five radish balady cultivars are shown in Table (12). The interaction was highly significant for all germination parameters.

**Table 12:** Effect of interaction between seed priming substances and salt levels on germination parameters overall radish genotypes.

Seed priming substances	Germination %**			Germination index**			Germination speed%**		
	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control
	NaCl	NaCl	(tap water)	NaCl	NaCl	(tap water)	ppm NaCl	ppm NaCl	(tap water)
H <sub>2</sub> O <sub>2</sub> (0.5%)	99.2abc#	99.4abc	98.8cd	98.8ab	99.2ab	98.8ab	99.3a	99.8a	99.9a
KNO <sub>3</sub> (3%)	99.1abc	96.5f	98.3de	97.2c	92.8e	97.6c	96.2a	92.9a	98.7a
NaCl (3%)	84.1h	66.4i	92.2g	67.6g	47.6h	77.6f	56.7c	40.9d	67.6b
Tab water	99.3abc	99.5abc	98.9bc	99.2ab	99.2ab	98.8b	99.8a	99.7a	99.4a
without	99.6ab	98.1e	99.7a	97.6d	94.4c	99.6a	96.6a	92.6a	99.7a
Seed priming substances	Seedling length (cm) **			Seedling dry matter %**					
	2000 ppm	4000 ppm	Control	2000 ppm	4000 ppm	Control			
	NaCl	ppm NaCl	(tap water)	ppm NaCl	ppm NaCl	(tap water)			
H <sub>2</sub> O <sub>2</sub> (0.5%)	5.43c	3.23f	7.23ab	5.5f	5.3g	7.4a			
KNO <sub>3</sub> (3%)	5.27cd	3.43ef	7.10b	5.2h	3.2i	6.9b			
NaCl (3%)	5.14d	2.56g	5.50c	5.9e	3.8k	5.5f			
Tab water	5.14d	3.50e	6.97b	5.1h	6.4c	6.1d			
without	5.22cd	3.31ef	7.43a	4.7i	4.2j	6.3c			

\*\* Interaction of such parameter is significant at level 1% of probability.

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability.

These results indicated that seed priming substances ranked as different as salt levels concerning such parameters. Bajehbaj (2010) revealed that the whole emergence of seedlings from both priming and non-priming seeds decreased with increasing NaCl salinity. Percentage of seed germination decreased with rising salinity levels in both primed and non-primed seeds. Elouaer and Hannachi (2012) showed that NaCl and KCl priming have improved germination parameters i.e., germination percentage, mean germination time, germination index, and coefficient of velocity and vigor index of safflower seed under saline conditions. Moghanibashi *et al.*, (2012) showed that primed seeds produced a higher germination index, germination rate days to 50% germination and germination index than non-primed seeds under all salinity levels. Pahoja *et al.*, (2013) indicated that hydro priming proved significantly better than osmo-priming (KNO<sub>3</sub>) under the wide range of salinity levels. Recently, Ashrafi *et al.*,

**Table 13:** Effect of interaction between radish genotypes and seed priming substances overall salt levels on germination parameters.

Seed priming substances	Germination %**					Germination index**					Germination speed%**				
	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut
<b>H<sub>2</sub>O<sub>2</sub> (0.5%)</b>	98.8bc	99.7ab	97.8d	100a	99.1abc	98.5cd	99.6abc	97.7de	98.7bcd	99.0abc	99.4a	99.7a	99.7a	99.3ab	99.8a
<b>KNO<sub>3</sub> (3%)</b>	94.4f	99.2abc	98.7c	99.6abc	97.6de	89.7i	97.4ef	96.6fg	87.7j	96.3g	90.6d	96.7bc	96.3bc	98.3ab	97.6abc
<b>NaCl (3%)</b>	60.5j	67.2i	8.77h	96.8e	92.1g	42.3n	46.1m	66.2l	99.7ab	78.8k	37.1h	36.3h	50.7g	80.7e	70.5f
<b>Tab water</b>	100a	99.5abc	99.1abc	99.8a	97.5de	99.8a	99.4abc	98.8abc	97.6de	97.1efg	99.7a	99.7a	99.5a	99.7a	99.1a
<b>without</b>	99.4abc	100a	99.7ab	99.5abc	96.8e	99.3abc	99.8a	94.0h	99.6abc	94.5h	99.8a	99.7a	89.5d	96.5bc	95.6c

Seed priming substances	Seedling length (cm)**					Seedling dry matter %**				
	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut	Alexandria	Kafr El-Sheikh	Elwady Elgadid	Gharbiya	Assiut
<b>H<sub>2</sub>O<sub>2</sub> (0.5%)</b>	40.0j	51.1defg	49.4fgh	55.5bcd	65.5a	7.5a	6.2d	4.7ij	6.5b	5.3fg
<b>KNO<sub>3</sub> (3%)</b>	42.7ij	45.5hi	56.6bc	63.3a	53.3cdef	4.9i	3.9k	6.4bc	4.9i	5.3fgh
<b>NaCl (3%)</b>	25.6m	34.1l	47.2gh	51.1defg	35.1kl	6.4bcd	3.4l	5.1h	4.9i	5.5ef
<b>Tab water</b>	46.6ghi	46.7ghi	52.2cdef	62.2a	50.5efg	5.3fg	5.6e	6.3cd	6.5bc	5.7e
<b>without</b>	38.8jkl	57.7b	52.2cdef	62.8a	54.6bcde	5.2gh	5.4fg	4.9i	5.4f	4.6j

\*\* Interaction of such parameter is significant at level 1% of probability.

#Values with the same alphabetical letters do not significantly differ from one another, using Duncan's multiple range test at 0.05 level of probability

(2015) concluded that hydro priming enhanced germination of all cultivars under both salt and drought stresses and non-stress conditions.

Concerning the effect of the interaction between seed priming treatments and studied genotypes, results illustrated in Table (13) showed significantly affect by the interaction between seed priming and genotypes in different parameters. These results are in good agreement with those reported by Moghanibashi *et al.*, (2012) and Moghadam and Mohammadi (2014).

### 3.2. PCR based on RAPD Analysis:

Characterization of plant with the use of molecular markers is an ideal way to conserve plant genetic resources. Molecular characterization helps to determine the breeding behavior of species, individual reproductive success and the existence of gene flow, that is, the movement of alleles within and between populations of the same or related species, and its consequences. RAPD can be suitable and efficient tool for genetic characterization of many plant species (Ghosh *et al.*, 2009), therefore in the present study the genetic diversity of some members of Brassicaceae family was determined using RAPD before the choice of a selected primer-marker system. It's required that RAPD primers should be evaluated statistically for its efficiency in assessing the polymorphic information. The calculation of statistically significant associations, the correlation among morphological and molecular parameters are the critical points within the final selection of markers.

Two primers for RAPD marker were screened for their ability to amplify the genomic DNA of the five studied turnip and radish cultivars (Photo1). Data were analyzed based on the comparison of the amplified fragments using gel documentation for each primer. If a fragment was present in a sample, it was designated as "1", if absent, it was designated as "0". If a fragment was present or absent in the genotype then absent or present in the others, it was called unique species-specific marker, but if a fragment was absent and present in more than one genotype, it was called polymorphic finally if the fragments was present in all genotypes, it was called monomorphic.

#### 3.2.1. Turnip

Data in Table 14 showed that total of 18 different fragments were obtained with the first primer, 7 fragments for Alexandria and Gharbiya, 8 for Kafr Elshikh and Assiut and 6 for Elwady Elgadid. 7 of the fragments were unique and 11 were polymorphic. The size of the amplified fragments varied from 195 to 1031 bp, RF varied from 0.642 to 0.184.

**Table 14:** Amplified DNAs fragments (AF) obtained for the five turnip genotypes using first RAPD primers.

AF	RF	MW	Genotypes					Polymorphism
			Alexandria	Kafr El-Sheikh	ELwady Elgadid	Gharbiya	Assiut	
1	0.184	1031	0	0	0	0	1	unique
2	0.239	813	0	0	1	0	0	unique
3	0.244	796	1	1	0	1	0	polymorphic
4	0.249	779	0	0	0	0	1	unique
5	0.284	673	0	1	1	1	0	polymorphic
6	0.289	659	1	0	0	0	0	unique
7	0.294	645	0	0	0	0	1	unique
8	0.348	517	1	1	0	0	0	polymorphic
9	0.353	507	0	0	0	1	1	polymorphic
10	0.388	443	1	1	1	0	0	polymorphic
11	0.393	434	0	0	0	1	1	polymorphic
12	0.448	355	0	0	1	0	1	polymorphic
13	0.453	349	1	1	0	1	0	polymorphic
14	0.512	284	0	1	0	0	0	unique
15	0.517	280	1	0	1	1	1	polymorphic
16	0.597	220	0	1	0	0	0	unique
17	0.637	198	1	1	1	0	0	polymorphic
18	0.642	195	0	0	0	1	1	polymorphic
<b>Detectable fragments</b>			7	8	6	7	8	

Data in Table 15 showed that total of 14 different fragments were obtained with the second primer, 4 fragments for Alexandria and Elwady Elgadiid, 5 for Gharbiya and Assiut and 3 for Kafr Elshikh. 9 of the fragments were unique and 5 were polymorphic. The size of the amplified fragments varied from 220 to 725 bp, RF varied from 0.658 to 0.312.

**Table 15:** Amplified DNAs fragments (AF) obtained for the five turnip genotypes using second RAPD primers.

AF	RF	MW	Genotypes					Polymorphism
			Alexandria	Kafr El-Sheikh	ELwady Elgadiid	Gharbiya	Assiut	
1	0.312	725	0	0	0	0	1	unique
2	0.332	671	0	0	0	1	0	unique
3	0.367	587	1	1	1	0	0	polymorphism
4	0.377	565	0	0	0	1	1	polymorphism
5	0.482	386	0	0	1	0	0	unique
6	0.508	354	1	1	0	0	0	polymorphism
7	0.513	348	0	0	0	1	1	polymorphism
8	0.538	320	1	0	0	0	0	unique
9	0.543	314	0	1	1	1	0	polymorphism
10	0.548	309	0	0	0	0	1	unique
11	0.593	267	1	0	0	0	0	unique
12	0.603	259	0	0	0	1	0	unique
13	0.608	255	0	0	0	0	1	unique
14	0.658	220	0	0	1	0	0	unique
<b>Detectable fragments</b>			4	3	4	5	5	

Table (18) showed the total AF obtained from DNAs of 5 studied turnip cultivars using two RAPD primers, it was ranked from 10 in Elwady Elgadiid to 13 in Assiut .

Amplified unique fragments obtained from DNAs of the five studied turnip cultivars using two RAPD primers were shown in Table (19) ranked from 2 in Kafr Elshikh and Gharbiya genotypes to 6 in Assiut genotype.

Table (20) showed a total of 32 RAPD fragments were amplified with the 2 used primers (18 in primer1 and 14 in primer2) at turnip cultivars, zero of them were common fragments (monomorphic), 16 of them showed to be polymorphic (50%) and other 16 showed to be unique fragments.

### 3.2.2. Radish

Data in Table 16 showed that total of 22 different fragments were obtained with the first primer, 7 fragments for Alexandria, Kafr Elshikh, and Gharbiya, 8 for Assiut, and 6 for Elwady Elgadiid. 13 of the fragments were unique 9 were polymorphic. The size of the amplified fragments varied from 237 to 1118 bp, RF varied from 0.659 to 0.118.

Data in Table (17) showed that total of 17 different fragments were obtained with the first primer, 5 fragments for Alexandria, Kafr Elshikh, and Elwady Elgadiid, 3 for Gharbiya and 7 for Assiut. 9 of the fragments were unique 8 were polymorphic. The size of the amplified fragments varied from 245 to 1374 bp, RF varied from 0.609 to 0.099.

Table (18) showed the total AF obtained from DNAs of 5 radish studied genotypes using two RAPD primers, it was ranked from 10 in Gharbiya to 15 in Assiut .

Amplified unique fragments obtained from DNAs of 5 radish studied genotypes using two RAPD primers were shown in Table (19) ranked from 2 in Gharbiya to 8 in Assiut genotype.

Table (20) showed a total of 39 RAPD fragments were amplified with the 2 used primers (22 in primer1 and 17 in primer2) at radish genotypes, zero of them were common fragments (monomorphic), 17 of them showed to be polymorphic (43.5%) and other 22 showed to be unique fragments.

**Table 16:** Amplified DNAs fragments (AF) obtained for the five radish genotypes using first RAPD primers.

AF	RF	MW	Genotypes					Polymorphism
			Alexandria	Kafr El-Sheikh	ELwady Elgaidid	Gharbiya	Assiut	
1	0.118	1118	1	1	0	1	1	polymorphic
2	0.199	888	0	0	1	0	1	polymorphic
3	0.204	876	0	0	0	1	0	unique
4	0.209	865	1	1	0	0	0	polymorphic
5	0.336	599	0	0	1	1	0	polymorphic
6	0.341	591	1	1	0	0	0	polymorphic
7	0.346	583	0	0	0	0	1	unique
8	0.408	489	0	0	0	0	1	unique
9	0.412	482	0	1	1	1	0	polymorphic
10	0.417	476	1	0	0	0	0	unique
11	0.512	362	0	0	0	0	1	unique
12	0.517	357	0	0	0	1	0	unique
13	0.521	352	0	0	1	0	0	unique
14	0.531	343	0	1	0	0	0	unique
15	0.54	334	1	0	0	0	0	unique
16	0.573	303	0	0	0	1	0	unique
17	0.578	299	0	1	1	0	1	polymorphic
18	0.588	291	1	0	0	0	0	unique
19	0.607	275	0	0	0	0	1	unique
20	0.611	272	0	0	1	1	0	polymorphic
21	0.616	268	1	1	0	0	0	polymorphic
22	0.659	237	0	0	0	0	1	unique
<b>Detectable fragments</b>			7	7	6	7	8	

**Table 17:** Amplified DNAs fragments (AF) obtained for the five radish genotypes using second RAPD primers.

AF	RF	MW	Genotypes					Polymorphism
			Alexandria	Kafr El-Sheikh	ELwady Elgaidid	Gharbiya	Assiut	
1	0.099	1374	0	0	0	1	1	polymorphic
2	0.103	1353	0	0	1	0	0	unique
3	0.108	1330	0	1	0	0	0	unique
4	0.245	823	0	0	0	1	1	polymorphic
5	0.250	807	0	0	1	0	0	unique
6	0.255	792	0	1	0	0	0	unique
7	0.258	787	1	0	0	0	0	unique
8	0.343	585	1	0	0	0	1	polymorphic
9	0.409	468	0	0	1	1	0	polymorphic
10	0.412	464	0	0	0	0	1	unique
11	0.416	458	1	1	0	0	0	polymorphic
12	0.506	337	0	0	1	0	1	polymorphic
13	0.515	330	1	1	0	0	0	polymorphic
14	0.541	304	0	0	0	0	1	unique
15	0.601	252	0	0	0	0	1	unique
16	0.603	250	0	0	1	0	0	unique
17	0.609	245	1	1	0	0	0	polymorphic
<b>Detectable fragments</b>			5	5	5	3	7	

**Table 18:** Amplified fragments (AF) obtained from the DNAs of the five turnip and radish genotypes using two RAPD primers.

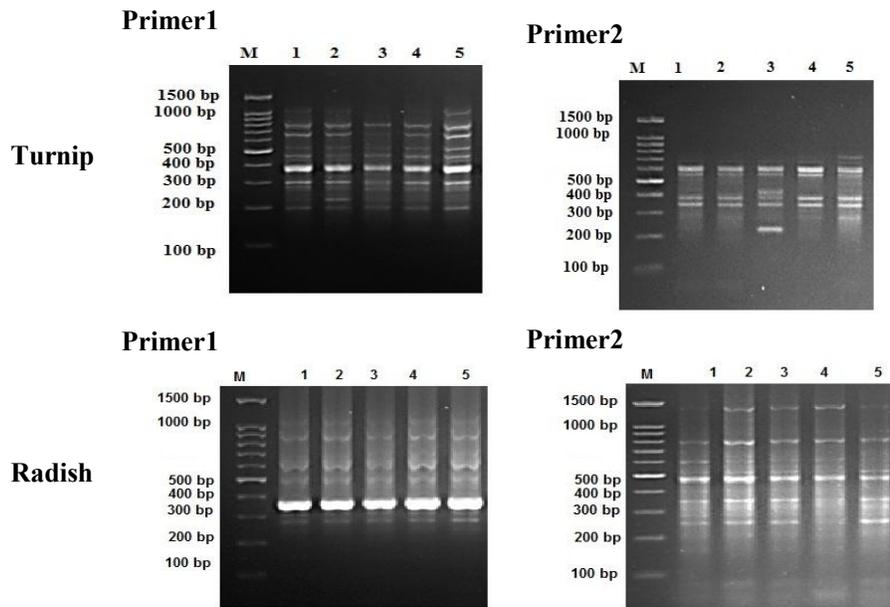
Species	RAPD Primer	Genotypes				
		Alexandria	Kafr El-Sheikh	ELwady Elgaidid	Gharbiya	Assiut
Turnip	OPA2	7	8	6	7	8
	OPA07	4	3	4	5	5
	<b>Total</b>	11	11	10	12	13
Radish	OPA2	7	7	6	7	8
	OPA07	5	5	5	3	7
	<b>Total</b>	12	12	11	10	15

**Table 19:** Amplified unique fragments obtained from the DNAs of the five turnip and radish genotypes using two RAPD primers.

Species	RAPD Primer	Genotypes				
		Alexandria	Kafr El-Sheikh	ELwady Elgadid	Gharbiya	Assiut
Turnip	OPA2	1	2	1	0	3
	OPA07	2	0	2	2	3
	<b>Total</b>	3	2	3	2	6
Radish	OPA2	3	1	1	3	5
	OPA07	1	2	3	0	3
	<b>Total</b>	4	3	4	3	8

**Table 20:** Amplified fragments (AF) obtained from the DNAs of five turnip and radish Species using two RAPD primers.

Species	RAPD Primer	PB	UB	TAF	P%
Turnip	OPA2	11	7	18	61%
	OPA07	5	9	14	35.7%
	<b>Total</b>	<b>16</b>	<b>16</b>	<b>32</b>	<b>50%</b>
Radish	OPA2	9	13	22	40.9%
	OPA07	8	9	17	47.1%
	<b>Total</b>	<b>17</b>	<b>22</b>	<b>39</b>	<b>43.5%</b>
<b>Total (Turnip + Radish)</b>		<b>33</b>	<b>38</b>	<b>71</b>	<b>46.5%</b>



**Photo 1:** RAPD banding patterns in the five turnip and radish genotypes accessions generated using two primers. (1, 2, 3, 4 and 5 for Alexandria, Kafr El-Sheikh, ELwady Elgadid, Gharbiya and Assiut, respectively).

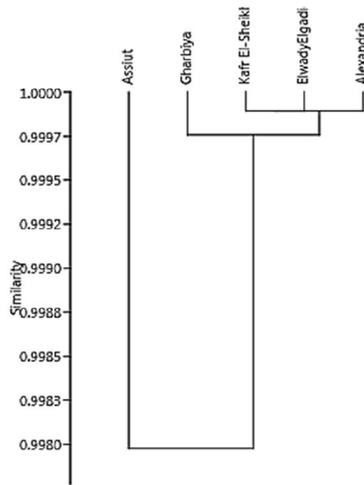
### 3.3. Clustering pattern

#### 3.3.1. Cluster diagram constructed using morphological markers

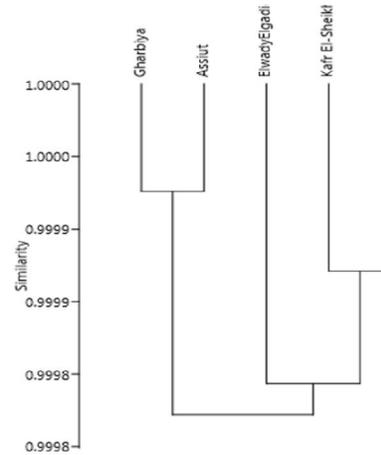
Cluster analysis is an important tool to describe and summarize the variation present between different plant species (Painkra *et al.*, 2018). It is obvious that after clustering, the species present in the same cluster would have similar characteristics, while those present in different clusters would be diverse. Cluster analysis using single linkage- correlation method depicting genetic similarity between five turnip and radish genotypes derived from sharing data of germination parameters among the genotypes overall seed priming and salt levels Fig. (1) divided the five turnip genotypes into two main groups and one sub-cluster. The first one contains Assiut genotype the second contain Gharbiya, Kafr Elshikh, Elwady Elgadid and Alexandria genotypes with sub-cluster contain Kafr Elshikh, Elwady Elgadid and Alexandria, (observe similarity values).

Regarding to radish genotypes cluster analysis Fig. (1) divided the genotypes into two main groups with one sub- cluster the first one contains Gharbiya and Assiut genotypes and the second one contains Elwady Elgadid, Kafr Elshikh and Alexandria genotypes with sub cluster Kafr Elshikh and Alexandri, (observe similarity values). There are some reports to show differences and correlation among different methods of diversity study along with their clustering patterns (Rahman *et al.*, 2011).

### Turnip



### Radish

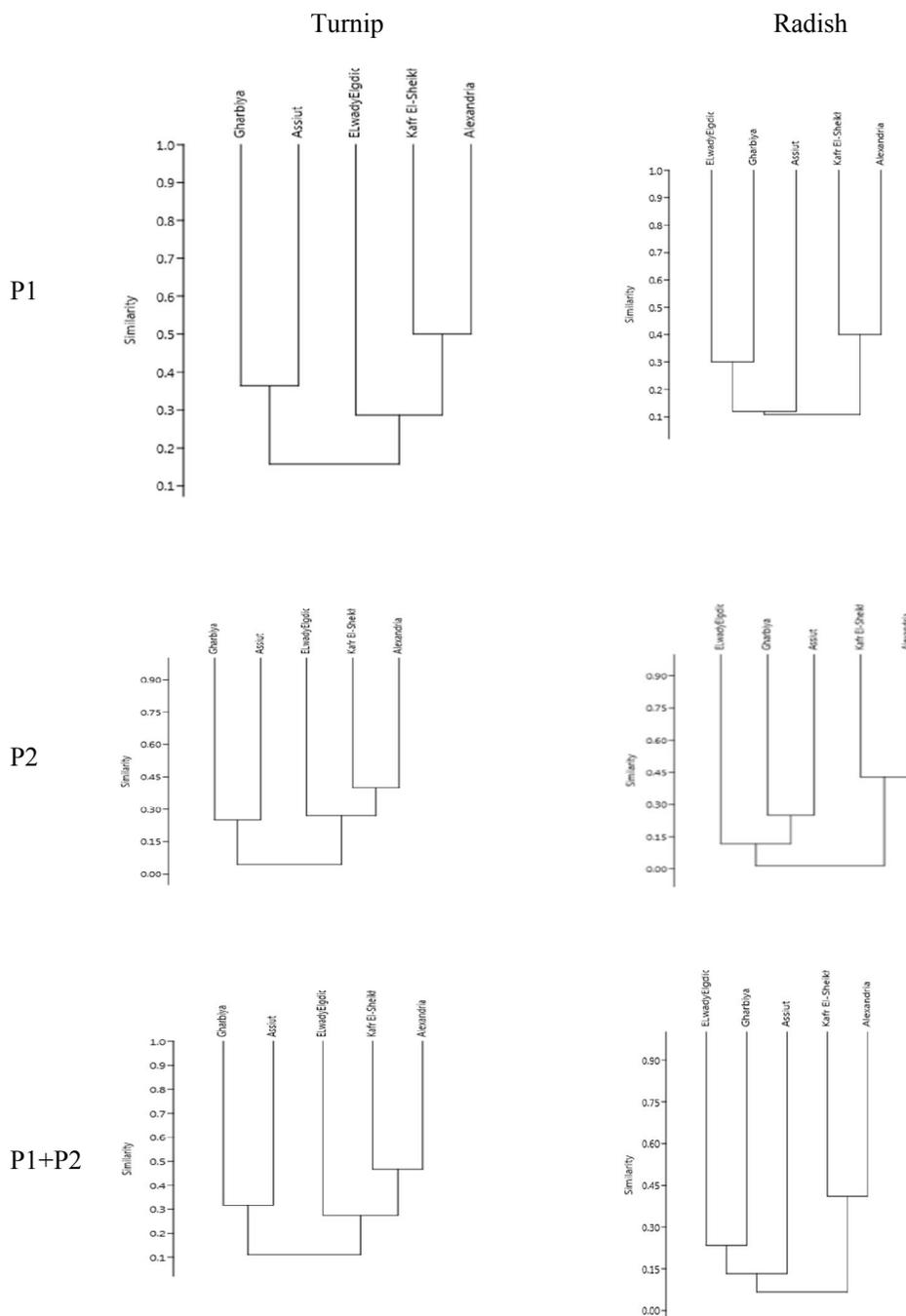


**Fig.1:** Cluster analysis using single linkage- correlation method depicting genetic similarity between five turnip and radish genotypes derived from sharing data of germination parameters among the genotypes overall seed priming and salt levels

### 3.3.2. Cluster diagram constructed using molecular markers:

Cluster analysis using the UPGMA method depicting genetic similarity (Jaccard's coefficient) between the five turnip genotypes derived from band sharing data of the 2 RAPD primers used divided the five genotypes into two main groups and one sub-clusters as shown in Fig. (2). the first group included Alexandria, Kafr Elshikh, and Elwady Elgadid which contain one sup order included Alexandria and Kafr Elshikh. The second group included Assiut and Gharbiya, there was no difference between clusters derived from the first, the second, and the both primers. It's important to evaluate the species diversity, which can add information on the breeding program. The study of species patterns and distribution is beneficial in understanding their system. Concerning clustering sites, it allows differentiating between those species that characterize individual groups, and those explain the similarity between species (Zhang 2017).

Regarding cluster analysis between the five radish genotypes derived from band sharing data of the 2 RAPD primers used it divided the five genotypes into two main groups and one sub-clusters as shown in Fig. (2). The first group included Alexandria and Kafr Elshikh, the second included Assiut, Gharbiya, and Elwady Elgadid, which contain one sub order included Gharbiya and Elwady Elgadid in the first and both primers used, meanwhile in the second primer the sub order included Assiut and Gharbiya, (observe similarity values) Fig. (2). Based on molecular data in the present study, it can be concluded that the molecular markers might be a good tool for studying genetic diversity.



**Fig. 2:** Cluster analysis using UPGMA method depicting genetic similarity (Jaccards coefficient) between the five turnip and radish genotypes derived from band sharing data of RAPD primers.

### 3.3.3. Genetic diversity among the studied Brassicaceae family members.

Regarding to study the genetic diversity among the studied Brassicaceae family members, a cluster analysis were obtained from sharing morphological and molecular data as shown in Fig.3 and 4, (observe similarity values).

The family Brassicaceae is one of the major groups of the plant kingdom, comprising of 340–360 genera and over 3,700 species distributed worldwide. Many species within the family are of great economic, agronomic and scientific importance. Some examples of these include the following: *Brassica napus* and *B. juncea* (oilseed crops); *B. rapa* (turnip, leaf vegetable); *B. oleracea* (cabbage, cauliflower, Kale, broccoli); *Raphanus sativus* (vegetable) and *Arabidopsis thaliana* (model plant)(Mark *et al.*, 2006). Although species from families Solanaceae and Brassicaceae are not typical halophytes, their seeds germinated well at a moderately elevated salt concentration.

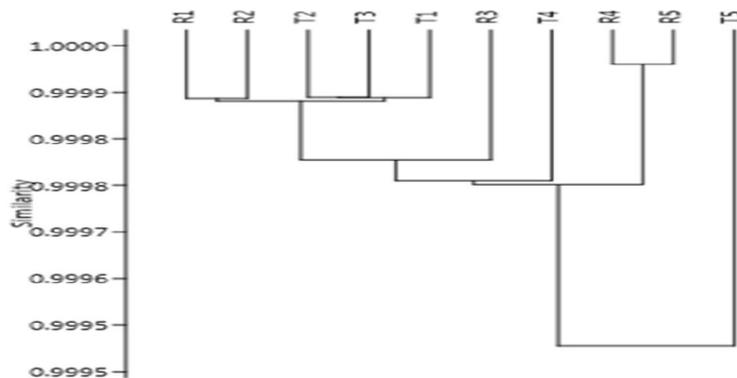
Early classifications depended on morphological comparison only, but because of extensive convergent evolution, these do not provide a reliable phylogeny. Although a substantial effort was made through molecular phylogenetic studies, the relationships within the Brassicaceae have not always been well resolved yet.

In this study, we introduce potential impacts of salinity on crop production of some Brassicaceae members, provide a reference for useful traits present in each of them and then we can concrete advice for structuring and optimizing introgression breeding programs.

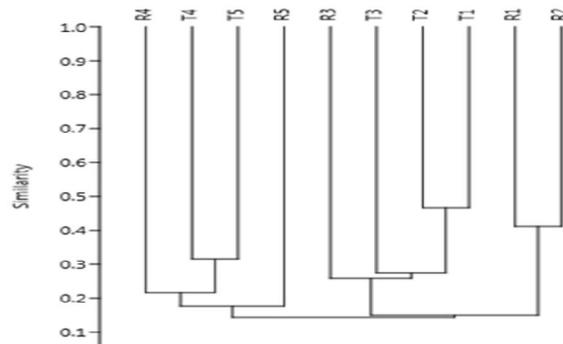
Success in transferring agronomically relevant traits between species depends on factors such as similarity between the source and target genomes. Regardless of the considerable difficulties involved in the use of wild relatives for crop improvement, this method offers a great deal of as-yet unexplored potential for the improvement of Brassica crops, and in improving crop resilience and salinity resistance.

RAPD can be suitable and efficient tool for genetic characterization of many plant species (Ghosh *et al.*, 2009), therefore in the present study the genetic diversity of the members of *Brassicaceae* family was determined using RAPD. Similarly (Lazoro and Aguinagalde, 1998) evaluated the genetic diversity in 29 populations of wild taxa of the *Brassica oleracea* L. group and two cultivars, using RAPDs as molecular markers. Nevertheless, before the selection of a specific primer-marker system, it is required that RAPD primers should be evaluated statistically for their efficiency in assessing the polymorphic information. The calculation of statistically significant associations, the correlation among morphological and molecular parameters are the critical points in the final selection of markers.

The two methods assessed high level of genetic variations. Based on combined results for morphological and molecular genetic diversity estimates, genotypes swapped among different clusters in different methods of clustering furthermore mono-genotypic clusters can be exploited to harness their unique features in breeding programs. Rahman *et al.* (2011), reported that genotypes also swapped from one cluster to another cluster among different methods and this pattern is somewhat irregular. These differences is not an indicator of the failure or limitation or weakness of the methods . These results may be due to the diversity at the molecular level, which may not reflect in the diversity at the morphological or physiological levels, as described by (Karhu *et al.*, 1996). Another possible reason for this variation in clustering might be the environmental influence and genotype environment interaction. Compared to morphological and physiological characteristics, the DNA genome provides direct comparison of genetic diversity at the DNA level, are phenotypically neutral and are not modified by environment and management practices (Messmer *et al.*, 1993). Morphological and physiological characters are the ultimate expression of molecular constitution of a variety where a number of biochemical processes is involved. So where a number of biochemical processes is involved. So different types of clustering in different methods is not unusual (Han-yong *et al.*, 2004). Molecular data improve or even allow the elucidation of phylogeny, and provide the basic knowledge for understanding taxonomy, domestication and evolution of plants .Information from molecular markers or DNA sequences offers a good basis for better conservation approaches. Modern molecular techniques have been developed in order to meet the demands of the horticulture industry. Population genetics, genetic linkage map and marker assisted selection techniques have significantly simplified the breeding procedures and overcome some of the agronomic, abiotic and biotic problems, which otherwise would not be achievable through conventional breeding methods.



**Fig. 3:** Cluster analysis using single linkage-correlation method depicting genetic similarity between five turnip and radish genotypes derived from combination of morphological data, letter T for turnip genotypes and letter R for radish genotypes, 1, 2,3,4 and 5 for Alexandria, Kafr El-Sheikh, ELwady Elgadid, Gharbiya and Assiut, respectively).



**Fig. 4:** Cluster analysis using UPGMA method depicting genetic similarity (Jaccards coefficient) between the five turnip and radish genotypes derived from combination of band sharing of RAPD data primers, letter T for turnip genotypes and letter R for radish genotypes, 1, 2,3,4 and 5 for Alexandria, Kafr El-Sheikh, ELwady Elgadid, Gharbiya and Assiut, respectively).

#### 4. Conclusion

Since salinity (whether in soil or irrigation water) is one of the obstacles to production. So, the present study was conducted to induce salinity tolerance in some native cultivars of turnip and radish through seed priming. In general, germination parameters were decreased as salt level increased up to 4000 ppm NaCl. It was found that the bad effect of salinity can be reduced on germination by using seed priming, generally, seed priming by H<sub>2</sub>O<sub>2</sub> (0.5%) or tap water gave the best results, meanwhile, seed priming by NaCl (3%) gave the worst results. On the other hand, the effect of salinity on germination parameters differed according to the cultivars of turnip and radish. This result indicates that the studied cultivars (for both turnip and radish) are genetically diverse. Therefore, cluster analysis, based on RAPD analysis, divided the five turnip cultivars into two main groups and one sub-cluster. The first one contains Assiut cultivar the second contain Gharbiya, Kaf Elshikh, Elwady Elgadid and Alexandria cultivar with sub-cluster contain Kafr Elshikh, Elwady Elgadid and Alexandria. Regarding to radish genotypes cluster analysis divided the cultivars into two main groups with one sub-cluster the first one contain Gharbiya and Assiut genotypes and the second one contain Elwady Elgadid, Kafr Elshikh and Alexandria genotypes with sub cluster Kafr Elshikh and Alexandria. Based on molecular data in the present study it can be concluded that the morphological and molecular markers could be a better tool for studying the genetic diversity. Accordingly, this study is a preliminary step towards the

development of new strains of turnip and radish tolerant to salinity, so it is recommended to complete this study.

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