GERMINATION PERFORMANCE OF SEQUENTIALLY HARVESTED TOMATO (LYCOPERSICON ESCULENTUM MILL.) SEED LOTS DURING SEED DEVELOPMENT UNDER SALT AND OSMOTIC STRESS

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ABSTRACT

This work concluded that tomato seeds harvested 70 days after anthesis showed the maximum germination not only under water but also salt stress. Seeds harvested earlier or later were more sensitive to stress at germinations. High Ki value of the lots indicated high resistance under stress conditions.

KEY WORDS : tomato, seed lots, germination, salt and osmotic stress

INTRODUCTION

Salinity stress is a major environmental constraint to irrigated agriculture in the arid, semi-arid and coastal regions. Flowers [1] estimated that about 40% of the world's irrigated agricultural lands could be considered as saline. Proper stand establishment and well-developed seedling production require rapid, uniform and complete germination. Once-over mechanical harvesting practices in most of the continuously flowering vegetable species including tomato are one of the determining factors of overall performance of seed lots, especially under stressful conditions. Seed lots, due to the once-over harvest are composed of a mixture of immature, mature and over-mature seeds extracted from early and late maturated fruits. Those seeds in the same lot which are less or over-maturated tend to show lower performance compared with fully maturated ones. This is accentuated by stressful sowing conditions such as low or high temperature, osmotic stress, saline soil and in turn, successful and timely stand establishment may not be achieved [2]. Salinity is one of the main constraining stress conditions that seeds encounter during germination in either direct sowing or transplanting production stages [3]. Salt tolerance is considered as a developmentally regulated stage specific phenomenon, and assessment of tolerance should evaluate separately for every developmental stage of the plant, i.e. seed germination, emergence, seedling, vegetative and reproductive growth [4]. However, the first exposure of the crop to salinity stress would occur at the germination stage in either soil surface (direct seeding) or seed-bed grown transplants in vegetables species, including tomato [5]. Seed development studies in tomato showed that the maximum seed quality assessed by a number of tests on serially harvested lots were obtained with seeds from fruits harvested in red-firm stage, 70 days after anthesis [6, 7]. Nevertheless, a seed batch can comprise seeds that were extracted from fruits harvested between pink (50 DAA) and overripened (90 DAA) stages due to the once-over mechanical harvests. Finding the uniformity, rapidity and germination percentage of serially harvested tomato seed lots under salt and osmotic stress might contribute to the efficiency of seedling production under stress conditions [8]. It will also help, determining the right fruit harvest for high quality seed in both direct sowing in saline soils or transplanting medium for seedling production.

The experiments presented here were undertaken to determine germination percentage and germination time of serially harvested tomato seed lots during seed development under salt and osmotic water stress.

MATERIALS AND METHODS

Tomato (Lycopersicon esculentum Mill.) plants of Rio Grande cultivar were grown in open-field conditions in the years of 1998 and 1999. The maximum and minimum temperatures were recorded as 39ºC and 12ºC during growing season, respectively. The rest of the plant husbandry conditions were as reported by [7]. Two hundred flowers during each year were tagged at full anthesis and fruits were harvested at 60 (red-firm), 70 (red-firm), 80 (red-soft) and 90 (overripened) days after anthesis (DAA) in 1998 and at 50 (pink), 60 (red-firm), 70 (red-firm) and 80 (red-soft) in 1999 Natural seed extraction was carried out by fermenting the slurry for 2 days at 25 ± 2ºC. Seed moisture content of fresh (after fermentation) and dried seeds, seed dry matter were determined according to [9].

To determine tolerance of tomato at germination to salt or osmotic stress during development, seeds were imbibed in distilled water, solutions of Polyethylene Glycol (PEG-6000) or NaCl. Germination percentage was calculated as average of four replications of 50 seeds. Seeds from each harvest and year, were placed on filter papers (Filtrak, Gmbh, Niederschlag 9303, Post Borenstein, Germany) moistened with 5.0 ml distilled water (control), NaCl (35, 70 and 140 mM), and PEG (–0.3, –0.6 and –0.9 MPa) in covered 9.0 cm diameter petri dishes and incubated at 25ºC in the dark for 14 days. Pomarsol whose active substance is Thiram as fungicide was added to the solutions at a concentration of 0.2% (w/v).

Germination was considered to have occurred when the radicle was 2 mm long and counts were made every day at the same time during the test. Mean time to germination was calculated according to [10].

Ki of the viability equation ($v = Ki - p/\sigma$) was proposed as objective and rational seed quality test (developed rapid ageing test) which shows the combined effect of all pre-storage factors (genetic and environment) on seed quality of the lot. In this equation v is probit percentage viability after p days in storage, σ is the standard deviation of the frequency of distribution of time and Ki is the seed lot constant. Ki varies between the lots and the higher the value is the better the quality (longer seed storage longevity, better performance under stress etc.). The procedure to determine the seed lot constant (Ki) of the viability equation as was proposed by Ellis and Roberts [10] for each lot as a seed quality assessment was as reported by [7]. Moisture contents of 1000 and 1400 seeds for each harvest in 1998 and 1999, respectively, were adjusted to 15% by placing them above water in closed containers at 20ºC until predetermined weights had been attained according to initial seed moisture and weight. The samples of 200 seeds in each species were stored in hermetically sealed aluminium foil-polyethylene pouches and stored at 50ºC. The samples were then removed from the store after 24, 48,72, 96, 120 h in 1998 and 8, 24, 36, 48, 72, 96, 120 in 1999 and germination tests (four replicates of 50 seeds) were conducted. Survival curves were constructed and linearized by plotting probit of percentage germination against time using SAS/Stat (Sas Institute Inc. Cary, NC., USA).Value of Ki were then (intercept on the y-axis) determined.

The differences between germination percentage means were compared with LSD (Least significant difference) test (P= 0.05) after angular transformation. The data shown in tables and figures are untransformed values.

RESULTS AND DISCUSSION

Mean seed dry weight of serially harvested seed lots did not change with time of harvest (P>0.05), but fruit colour changed from pink (50 DAA) to red soft (80 DAA) or overripened (90 DAA) (Table 1). Ki of the viability equation increased with the advancement of maturity and reached a maximum at seeds harvested 70 DAA with 2.61 and 2.54 in 1998 and 1999, respectively and decreased again at later harvest times.

Some of the earlier studies showed that seeds harvested from red-firm fruits or 70 DAA attained the maximum seed quality traits assessed by germination, emergence, seedling dry weight and seed storage longevity [11, 6].

The response of plants to saline conditions relies on a number of different factors, including plant species, stage of development, growth environment, concentration and length of time to salt exposure. Although suppression of growth and yield of tomato due to salinity has been reported [12], its effect on germination, particularly of differentially matured lots is not known [13].

Percentage of germination of tomato seeds harvested 60, 70 and 80 DAA was higher than 90% at all concentrations of NaCl tested in 1998 seed lots. Seeds harvested at 90 DAA from overripened fruits, germination gradually declined from 92.5% in control, to 90.0, 82.5 and 76.5% at 35, 70 and 140 mM, respectively (Figure1). However, all seed lots harvested in 1999 had higher germination rates than 90% at 35 and 75 mM NaCl concentrations, but seeds harvested at 80 DAA had 88.5% of germination at 140 mM NaCl (Figure 2.). But only differences among the lots was not statistically significant (P<0.05).

In our study inhibition of the germination of tomato seeds by NaCl was greater at 140 mM and in later harvests (80 and 90 DAA). The germination of cucumber seeds decreased under 90% at NaCl concentration of 150 mM [3]. However, germination of some Lycopersicon esculentum cultivars decreased as low as 75% at 100 mM [14]. This shows that stress tolerance is not only genetically controlled trait, but also controlled by the developmental stage of the seed [15]. The tolerance of seed at germination to salinity is a measure of the seed ability to withstand the effects of high concentration of soluble salts in the medium. The seeds then, should generate required osmotic potential to improve the water status of the embryo, permitting protrusion of the radicle [16]. Results in this experiment proved that seeds harvested 70 DAA in both years are more capable of improving water status of the embryo than earlier (50 DAA) or later (80 and 90 DAA) harvest stages, since they showed maximum germination at all NaCl concentrations (Figures1 and 2). In agreement with these conclusions, [17] also found that unmature or overmature harvested seeds of cabbage and rape were more sensitive to externally applied ABA or low water potential stresses than optimally mature seeds.

Salinity influenced germination time more dramatically than germination percentage. Despite the fact that germination percentage was not significantly affected (P>0.05), the difference in mean germination times between seeds germinated at 35 mM and 140 mM was significant at about 3 days (Figures 1b and 2b). It means that the higher the salt concentration the longer the time to germination. Within all seed lots, the shortest time to germination was observed for the seeds harvested 70 DAA being 3.12, 3.87 and 6.55 in 1998, and 3.14, 3.75, 6.77 in 1999 at 35, 70 and 140 mM NaCl concentrations, respectively (Figures 1b and 2b).

This clearly shows that, although viability is a well accepted component of seed quality, mean time to germination as a seed vigour trait, can be more discriminative in ranking the seed lots regarding performance under stress as reported also by [18]. Therefore, sensitivity to stress manifests itself through mean time to germination, and through seed vigour, rather than through germination percentage in this experiment. In agreement with that conclusion, seed vigour features such as seedling growth, dry weight and emergence in carrot [19] and cucumber [3] have been reported to be more sensitive to salinity comparing to seed germination.

Germination percentages under low water potential showed that none of seeds of any harvest germinated at –0.9 MPa. At –0.6 MPa, germination was varying between 8.50 and 22.50% among the lots. Seeds harvested 60 DAA gave the highest germination percentages at –0.3 MPa among all lots, being 97 and 96% in 1998 and 1999, respectively (Table 2). In another study, aqueous solutions of NaCl used in this experiment gave osmotic potentials of –0.16, –0.33, and –0.66 MPa (35, 70, and 140 mM, respectively) [20]. Tomato seed germination was more

Table 1. Changes in seed dry weight, fruit colour and Ki (seed lot constant) of tomato seed lots harvested in different days after anthesis (DAA) in the years of 1998 and 1999.

Year	DAA	Seed dry weight (mg/seed)	Fruit colour	Ki
1998	60	3.13	Red-firm	2.07
	70	3.06	Red-firm	2.61
	80	3.30	Red-soft	2.22
	90	3.23	Overripened	1.72
1999	50	2.90	Pink	1.92
	60	2.84	Red-firm	2.22
	70	2.85	Red-firm	2.54
	80	2.77	Red-soft	2.14

Table 2. Germination percentages and mean time to germination (MGT) (days) of the seed lots at –0.3, -0.6 and –0.9 MPa (Mega pascal) of PEG solution. Mean time to germination was calculated only for – 0.3 MPa since very low germination was recorded for other concentrations.

Year	DAA	MPa –0.3 Germ. (%).	MPa –0.3 M.G.T. (day).	MPa –0.6 Germ. (%).	MPa –0.9 Germ. (%).
1998	60	97.5	4.55	13.5	0
	70	84.7	4.28	16.7	0
	80	91.0	4.52	11.5	0
	90	82.5	4.54	22.5	0
LSD	-	6.8	0.4	-	-
1999	50	92.0	6.09	8.5	0
	60	96.0	5.02	15.5	0
	70	91.5	4.09	15.5	0
	80	91.0	4.09	13.0	0
LSD	-	7.8	0.4	-	-

dramatically influenced by osmotic water stress than salt stress despite osmotic potentials were similar about –0.6 MPa. However, it was found for lettuce that toxicity effect of NaCI rather occurs in seedling growth stage subsequent to radicle emergence [20]. Passam [3] also found that despite seed germination is not significantly different between 50 and 200 mM of NaCI, toxicity (necrotic areas on leaves) occurred at concentrations of 50 and above in cucumber seedlings. It appears to be in the present work that osmotic stress rather then toxicity is the main constraint during germination (at least up to 140 mM) which restricts water to reach threshold level for germination in tomato.

Similar to our findings, Welbaum [21] in muskmelon and Still [22] in broccoli reported that germination under low water potential was influenced by maturity stages. [10] proposed that Ki, seed lot constant of the viability equation can be considered as a good indicator of the seed quality due to the its good correlation with other important characteristics of a seed lot. The result of this experiment showed that seeds harvested 70 DAA, not only had the maximum Ki value but also maximum germination and minimum time to germination at all NaCl concentrations except for two cases (140 mM total germination 1998, 70 mM mean time to germination 1999) (Table 1, Figures1a and b and 2a and b). Therefore, Ki appears to be associated with germination performance of a seed lot under salt stress.

Concerning the determination of the right time of seed harvest in tomato, seeds extracted from red-firm 70 DAA had maximum germination rates, emergence and longevity at stress temperatures [7]. Moreover,

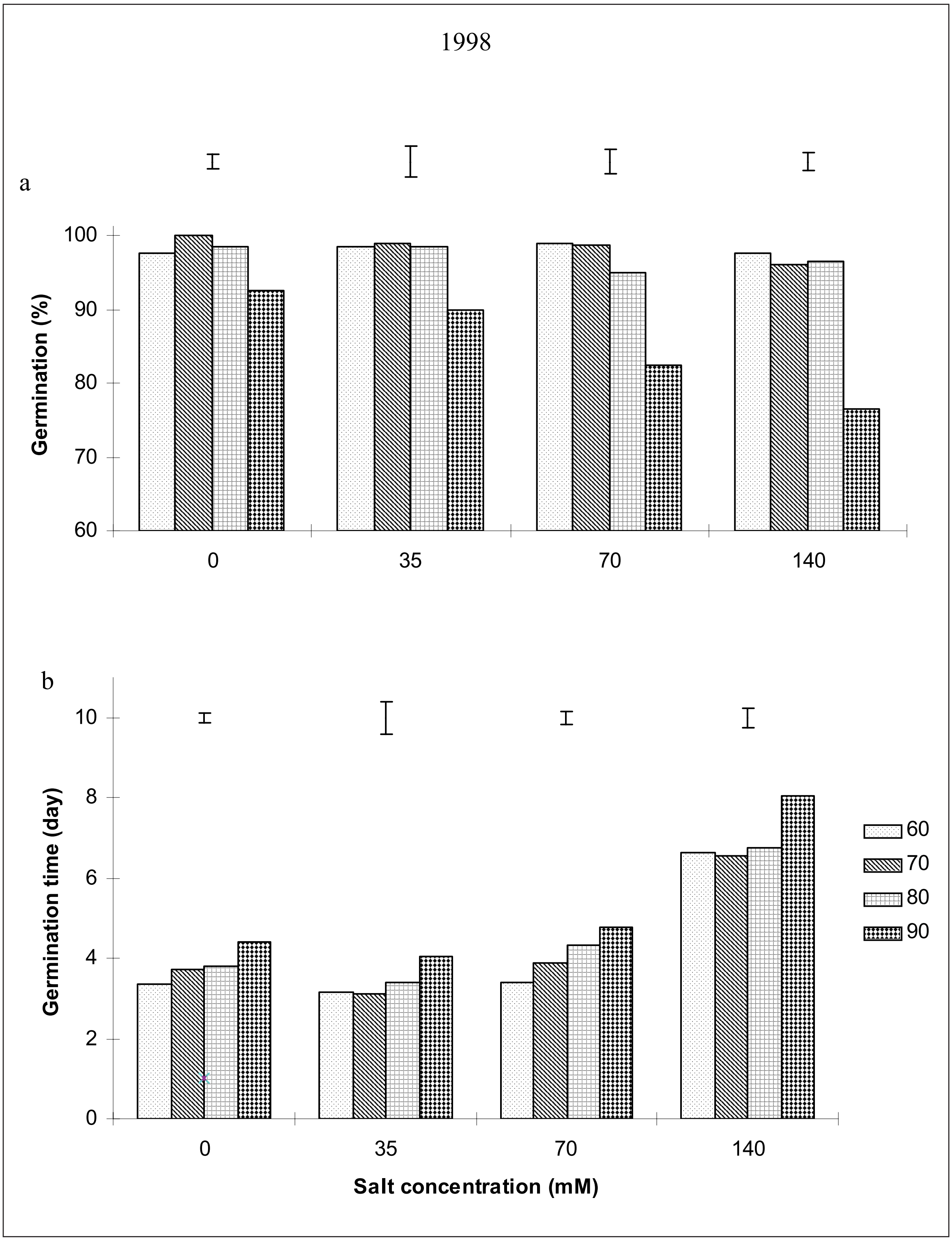

Fig. 1. Germination percentage (a) and mean time to germination (b) of tomato seed lots harvested 60, 70, 80 and 90 days after anthesis (DAA) in 1998 at salt concentrations of 35, 70 and 140 mM NaCl. LSD(P 0.05) values are shown as bars.

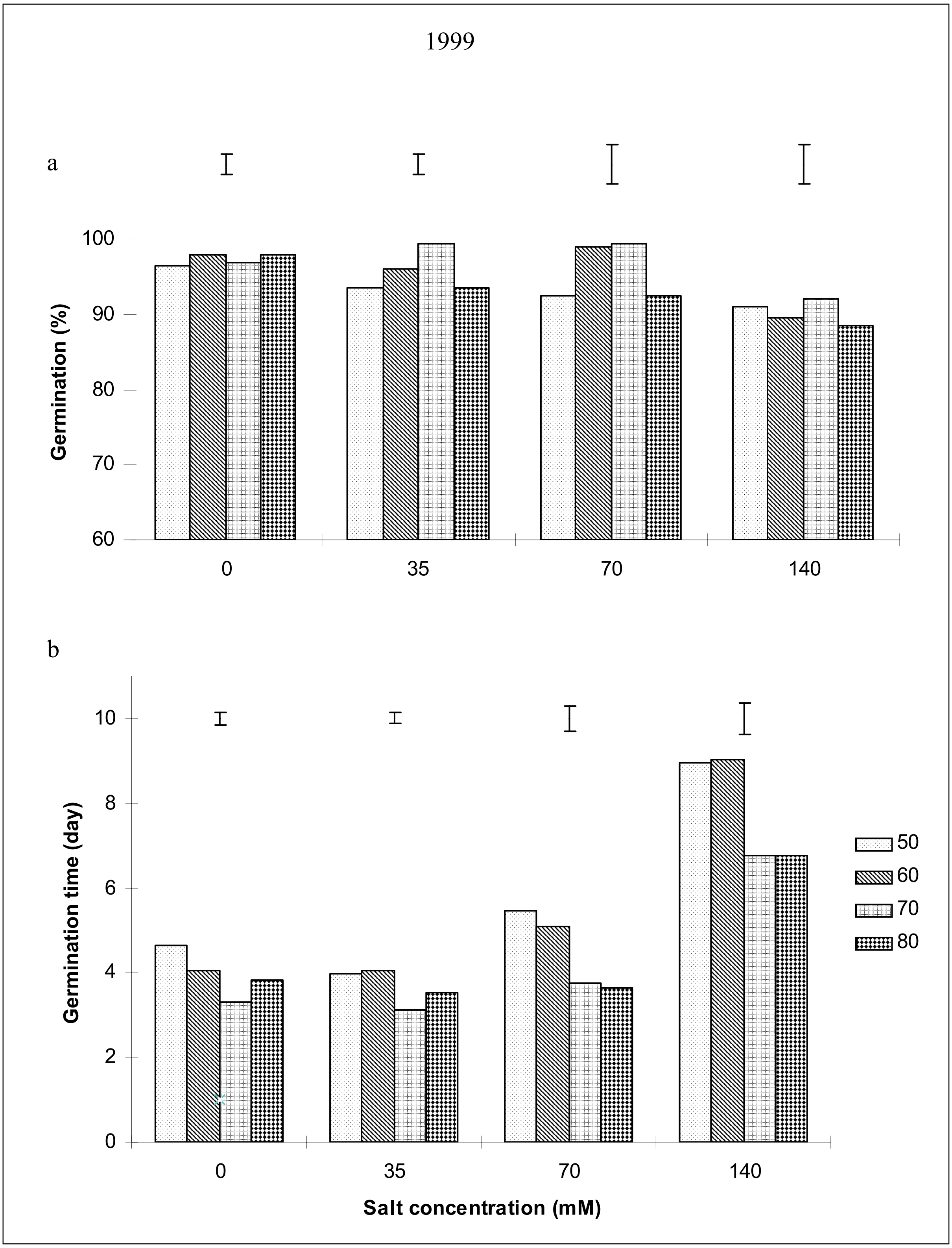

Fig. 2. Germination percentage (a) and mean time to germination (b) of tomato seed lots harvested 50, 60,70 and 80 DAA in 1999 at salt concentrations of 35, 70 and 140 mM NaCl. LSD (P 0.05) values are shown as bars.

this experiment showed that they also had maximum germination performance under salt stress. Tomato seeds should be harvested when fruits are red-firm, (about 70 DAA) in order to get maximum germination efficiency and stand establishment, which is a concern in saline soils. Earlier or later seed harvests will reduce salt stress tolerance.

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