RESEARCH ARTICLE

EFFECT OF SALINITY ON SEEDLING GROWTH OF CITRUS ROOTSTOCKS

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Abstract: The experiment was carried out at screen house of the Department of Horticulture, CCS Haryana Agricultural University, Hisar for the two consecutive years during the year 2018-19 and 2019-20 to extrapolate the effect of rootstock and salinity on seedling parameters of nine different citrus rootstocks. Seedling height, stem diameter, number of leaves per plant and number of seedlings emerged per seed were adversely affected when subjected to soil salt stress from control (0.07 dS/m) to 7 dS m^{-1}) Among all rootstocks, Rangpur lime, followed by Volkamer lemon and Cleopatra mandarin were found better with relatively less reduction at 7 dS m^{-1} over control, whereas Pectinifera, followed by NRCC-4 and Alemow were found inferior which showedrelatively high reduction at 7 dS m^{-1}over control in respect of seedling height, stem diameter and number of leaves per plant at seedling stage.

Keywords: Citrus, Rootstocks, Salinity, Seedling growth

INTRODUCTION

Citrus is one of the most commercially grown fruit crops in the world and is third most important fruit crop of India accounting 12.4 per cent of total fruit production. It is grown predominantly in tropical and subtropical climates and is found to be the most salt-sensitive crop, thereby becoming more vulnerable to drought and salinity problems (Simpson *et al*., 2015; Sahin-Çevik *et al*., 2020). The damage by viral infections can only be checked by delivering adequate and precise information about viral diseases to the Kinnow fruit growers, because if they are not managed in time, they can even wipe out the whole citrus industry (Dixit and Mohan, 2012). Palchoudhury *et. al*. (2019) reported that potting mixture containing soil, sand and vermiculite is very effective for survival and healthy growth of Kagzilime plant. Though, citrus is highly salt sensitive crop, disparity in tolerance do exist among species (Mass, 1993). Citrus genotypes can be classified as relatively tolerant and more sensitive to salinity owing to their ability to restrict chloride ions to roots (Lopez-Climent *et al*., 2008).The diversification of citrus rootstocks have anenormous importance due to their influence on tree vigour, water relations, production, hormonal balance, mineral nutrition, quality and tolerance to abiotic and biotic agents that reduces the risk of suffering enormous losses (Bowman and Joubert, 2020). No promising results are obtained so far concerning new citrus cultivars and rootstocks tolerant to salinity (Helaly and Hanan, 2011), which is probably due to the complex mechanisms involving salinity tolerance in plants (Egamberdieva and Lugtenberg, 2014). In view of the above facts, the experiment was envisaged to study the effect of rootstock and salinity on seedling parameters of different citrus rootstocks.

MATERIALS AND METHODS

The present investigation was conducted at CCS HAU, Hisar during 2018-20. Hisar has a typical semi-arid climate with hot and dry summer and extremely cold winter. The soil was collected from the sand dunes from Balasmand village in Hisar district. The collected soil samples were passed through 2 mm sieve and subjected to mechanical and chemical analysis. The soil was found to be sandy in texture, low in organic carbon, medium in available N and P with fairly alkaline reaction and saturation capacity of 25%. The crop was raised in plastic pots filled with 10 kg of dune sand and was supplied with Hoagland nutrient solution at regular intervals. The salinity was developed in the soil with the help of artificial waters of different ionic compositions as specified in Table 1. The required amount of chloride and sulphate salts of Na^+, Ca^{2+} and Mg^{2+} were added through NaCl, $CaCl_2.2H_2O$, $MgCl_2.6H_2O$ and $MgSO_4.7\ H_2O$. The salts were taken in a 100-litre beaker and dissolved in water and final volume was made to 75 litres separately for each salinity level. From this solution 2.6 1/pot was added to pots containing 10 kg of soil for maintaining their respective salinity level. After drying, this soil was thoroughly mixed.The research comprised of the nine different rootstocks and five chloride dominated salinity levels.

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Table 1. Composition of different ions for preparing chloride dominated saline water

EC_e level (dSm^{-1})	Total dissolved salts (TDS)	Na^+	Ca^{2+}	Mg^{2+}	Cl^-	SO_4^{2-}
		me/l				
2.5	30.50	15.25	3.81	11.43	21.35	9.15
4.0	50.0	25.0	6.25	18.75	35.0	15.0
5.5	66.50	33.25	8.31	24.93	46.55	19.95
7.0	86.0	43.0	10.75	32.24	60.20	25.80

The seeds of the nine citrus rootstocks *viz*., Rough lemon (*Citrus jambhiri* Lush.), Cleopatra mandarin (*Citrus reshni* Tanaka), Pectinifera (*Citrus depressa* Hayata), Rangpur lime (*Citrus limonia* Osbeck), Alemow (*Citrus macrophylla* Wester), Volkamer lemon (*Citrus volkameriana*), NRCC-3, NRCC-4 and CRH-12 were collected from a single treefor each rootstock from CCRI, Nagpur; CEF, Mangiana and CCS HAU, Hisar. Seeds were extracted from fruits, washed with water and air dried under shade and treated with Bavistin @ 2g/100g seeds. These weresown at a depth of one cm. After sowing, the seeds were covered with the soil. The seeds were sown in three replications with 10seeds/replicationin the pots with salinity level 0.07 (control), 2.5, 4.0 and 7.0 dS m^{-1}. These treatments were laid in Completely Randomized Design (CRD). Observations were recorded on seedling height (cm), stem diameter (mm), number of leaves per plant and number of seedlings emerged per seed.

RESULTS AND DISCUSSION

Salinity level and rootstocks significantly influenced the seed germination parameters, namely seedling height (cm), stem diameter (mm), number of leaves per plant and number of seedlings per seed.The data pertaining to seedling height presented in Table 2showed that rootstocks differed significantly among each other in both the years of study. The maximum (21.02 cm) seedling height was observed in Volkamer lemon, which was found statistically at par (20.97, 20.91 and 19.16 cm)with Rangpur lime, Alemow and Rough lemon, respectively, whereas, the minimum (15.01cm) seedling height was recorded in Pectinifera, which was at par (16.60 cm)with Cleopatra mandarin, irrespective of the salinity level. However, within rootstocks, NRCC-3, NRCC-4, CRH-12 and Rough lemon; Cleopatra mandarin and NRCC-4 were found statistically at par with each other.Seedling height was decreased significantly with gradual increase in the salinity levels. The maximum seedling height (22.37 cm) was observed at control (0.07 dS/m), which was significantly at par (22.07 and 20.62 cm) with that at 2.5 and 4.0 dS m^{-1}, while this was found minimum (11.83cm) at 7 dS m^{-1}, irrespective of the rootstock. Also, among various salinity levels, seedling height at control, 2.5 and 4 dS/m were found statistically at par with each other.The interaction between rootstocks and salinity levels was found significant in both the years. The maximum (25.00 cm)seedling height was observed in Alemow at control in the year (2018-19), closely followed by Volkamer lemon, Rangpur lime, while the minimum (8.33)seedling height was observed in Pectinifera, at par with NRCC-4 and Cleopatra mandarin at 7 dS m^{-1}. Theminimum reduction in seedling height was observed in Rangpur lime (39.72%), followed by Volkamer lemon (43.24%) and Cleopatra mandarin (43.86%), whereas, the maximum reduction was observed in Pectinifera (55.36 %), followed by NRCC-4 (54.55%) at 7 dS m^{-1}over control in the year 2018-19. Similar trend was noticed in seedling height in the next year (2019-20) among each salinity level for all the rootstocks.The stem diameter was significantly influenced by salinity level and rootstocks in both the years (Table 3). In general, with an increase in salinity level, there was an increase in reduction over control in stem diameter. Among different rootstocks, the maximum (2.50 mm) stem diameter was observed in Volkamer lemon, which was statistically at par with Rough lemon, Cleopatra mandarin, Rangpur lime and NRCC-3 (2.48, 2.42, 2.37 and 2.35 mm), respectively, whereas, the minimum stem diameter (2.12 mm) was recorded in CRH-12, which was found at par with Alemow, Pectinifera, NRCC-4, NRCC-3 and Rangpur lime (2.14, 2.16, 2.21, 2.35 and 2.37 mm), respectively, irrespective of the salinity level. Among various salinity levels, maximum (2.49 mm) stem diameter was recorded at control, which was at par with that at 2.5 and 4 dS m^{-1}(2.44 and 2.36 mm), respectively, while, this was found minimum (2.01 mm) at 7 dS m^{-1}, irrespective of the rootstock in the year 2018-19. Data revealed that the interaction between rootstocks and salinity levels was significant in both the years.Among different rootstocks, the maximum stem diameter was recorded in Volkamer lemon (2.67 mm), which was found significantly at par with all other rootstocks at control, except Alemow and CRH-12, whereas, the minimum stem diameter was recorded in Pectinifera (1.72 mm), which was at par with Alemow, NRCC-4 and CRH-12 (1.79, 1.87 and 1.87 mm), respectively at 7 dS m^{-1}in the year 2018-19. The maximum reduction (2.40 to 1.72 mm; 28.3%) was recorded in Pectinifera, followed by Alemow

(2.35 to 1.79 mm; 23.8%) and NRCC-4 (2.44 to 1.87 cm; 23.4%), whereas, the minimum reduction of stem diameter (2.52 to 2.15 mm; 14.7%) was recorded in Rangpur lime, followed by Volkamer lemon (2.67 to 2.50 mm; 14.9%) and Cleopatra mandarin (2.57 to 2.18 mm; 15.1%), as the salinity levels increased from control to 7 dS/m in the year 2018-19. Similar results of stem diameter were observed in the year 2019-20 among each salinity level for all the rootstocks. This may be due to differential response of rootstock to salinity. Reduction in growth parameters at increasing salinity levels can, in some cases, be attributed to salinity-induced adverse change in leaf water relations reducing photosynthesis, dehydration of proteins and protoplasm to a lower extent (Nieves *et al*., 1991). The decreased growth might also be due to osmotic effect of salt on root and toxic effect of accumulated ions in the plant tissues (Lea-Cox and Syversten, 1993; Storey,1995). The results are in accordance with the findings of Adams *et al*. (2019) who perceived an average reduction in height (20.2%) and stem diameter (5.2%) of plants when treated with 150 mM NaCl than the control plants in 'US-942' citrus rootstock. In addition, 30% reduction was observed in height among US942 and X639 rootstocks, whereas, FA No. 5, Carrizo citrange and US897 showed a reduction of 35, 38 and 47%, respectively at the highest salt concentration (75 mM) when compared with the control treatment (Aparicio-Durán, 2021). Moreover, the shoot growth of Volkamer lemon, Rough lemon and Sour orange decreased to over 70% of that in the control at 135 mM NaCl (Alam *et al*., 2020).

With respect to the number of leaves per plant, a significant decrease with an increase in salinity level from control to 7 dS m^{-1} in both the years was observed (Table 4).Among different rootstocks, the maximum number of leaves per plant (24.77) was observed in Rangpur lime followed by Volkamer lemon (21.23) whereas, the minimum number of leaves per plant was recorded in NRCC-4 (13.90), which was statistically at par with Alemow (15.05), irrespective of the salinity level. However, within rootstocks, Rough lemon, Cleopatra mandarin and Volkamer lemon; Pectinifera, NRCC-3 and CRH-12 were found statistically at par with each other.Among various salinity levels, number of leaves per plant was found maximum (20.89) at control, which was significantly at par with that at 2.5 dS m^{-1}(20.15), after that there was gradual decrease with salinity and finally it was minimum (15.69) at 7 dS m^{-1}, irrespective of the rootstock. The interaction between rootstocks and salinity levels was found significant in both the years. The maximum number of leaves per plant was observed in Rangpur lime (27.33) at control, which was at par with Rangpur lime at 2.5 and 4.0 dS $m^{-1}$and also with Rough lemon at control and 2.5 dS m^{-1}, while the minimum number of leaves per plant was recorded in NRCC-4 (11.67), closely followed by Alemow. Thickened and fewer number of leaves are some common morphological symptoms of plants in saline medium. A progressive decline in leaf number was observed under salt stress. These results confirm the findings of Anjum *et al*. (2000) who reported that as salinity level of the soil was increased from EC_e 1.65 to 8 dSm^{-1}, number of leaves per plant were progressively decreased with least effect on the performance of Cleopatra mandarin as compared to Troyer citrange, the most affected rootstock, while Red Rough lemon, Bitter sweet orange and Volkameriana were in between at all the EC_e levels.

The number of seedlings per seed decreased significantly with an increase in salinity levels from control to 7 dS/m in both the years (Table 5). Among different rootstocks, the maximum number of seedlings per seed (1.70) was observed in Cleopatra mandarin, which was at par with Pectinifera (1.59), whereas, the minimum number of seedlings per seed (1.02) was recorded in Rangpur lime, irrespective of the salinity level. However, Rough lemon, Alemow, Volkamer lemon, NRCC-4 and CRH-12; Pectinifera, Alemow and Volkamer lemon were found statistically at par with each other in the year 2018-19. Among various salinity levels, number of seedlings emerged per seed was found maximum (1.54) at control (0.07 dS m^{-1}), at par (1.51 and 1.37) with that at 2.5 dS m^{-1} and 4 dS m^{-1}, respectively, while, minimum (1.11) at 7 dS/m irrespective of the rootstock. However, number of seedlings emerged per seed were found at par with each other with gradual increase in salinity. The data revealed that the interaction between rootstocks and salinity levels was also found significant in both the years. The number of seedlings per seed was found maximum in Cleopatra mandarin (2.08), at par with Pectinifera (1.83) at control and the minimum inRangpur lime and NRCC-3 and CRH-12 (1.00) at 7 dS m^{-1}in the year 2018-19. Similar trend was also noticed in the number of seedlings emerged per seed in the succeeding year. This might be due to different genetic behaviour and degree of polyembryony in the species and their sensitivity to salinity. The results are in accordance with the findings of Murkute *et al*. (2010) who recorded 3.2 and 3.7 number of shoots/ explants in *C. jambhiri* and *C. karna* respectively, which got reduced to 1.06 and 1.02 number of shoots/ explants under highest salinity level (100 mM).

Table 2. Effect of different salinity levels on seedling height (cm) of citrus rootstocks

Rootstocks	Salinity level (dS m^{-1})											
	2018-19						2019-20					
	0.07 (Control)	2.5	4.0	5.5	7.0	Mean	0.07 (Control)	2.5	4.0	5.5	7.0	Mean
Rough lemon	22.67	22.50	20.95	17.33	12.33	19.16	19.72	19.30	18.02	14.95	11.72	16.74
Cleopatra mandarin	19.00	19.00	18.67	15.67	10.67	16.60	16.67	16.21	15.44	14.31	10.14	14.55
Pectinifera	18.67	18.33	16.73	13.00	8.33	15.01	16.11	15.55	13.85	11.76	7.93	13.04
Rangpur lime	24.33	24.10	22.73	19.00	14.67	20.97	21.12	20.72	20.12	17.58	13.94	18.70
Alemow	25.00	24.50	22.87	19.67	12.50	20.91	21.50	20.85	19.64	18.21	11.89	18.42
Volkamer lemon	24.67	24.33	22.87	19.25	14.00	21.02	21.38	21.08	19.57	16.61	13.32	18.39
NRCC-3	22.67	22.33	20.67	16.67	11.67	18.80	19.57	18.89	17.25	14.30	11.09	16.22
NRCC-4	22.00	21.50	19.85	15.33	10.00	17.74	18.87	18.10	16.53	13.61	9.75	15.37
CRH-12	22.33	22.00	20.25	17.25	12.33	18.83	19.33	19.00	17.25	14.81	11.73	16.43
Mean	22.37	22.07	20.62	17.02	11.83		19.36	18.86	17.52	15.13	11.28	
C.D. at 5%	**Rootstock = 1.93 Salinity = 1.89 Rootstock x Salinity = 3.12**						**Rootstock = 1.88 Salinity = 1.84 Rootstock x Salinity = 3.05**					

Table 3. Effect of different salinity levels on stem diameter (mm) of citrus rootstocks

Rootstocks	Salinity level (dS m^{-1})											
	2018-19						2019-20					
	0.07 (Control)	2.5	4.0	5.5	7.0	Mean	0.07 (Control)	2.5	4.0	5.5	7.0	Mean
Rough lemon	2.64	2.62	2.53	2.39	2.21	2.48	2.61	2.58	2.50	2.36	2.18	2.45
Cleopatra mandarin	2.57	2.53	2.49	2.35	2.18	2.42	2.55	2.54	2.47	2.33	2.16	2.41
Pectinifera	2.40	2.35	2.28	2.05	1.72	2.16	2.33	2.30	2.24	1.99	1.67	2.11
Rangpur lime	2.52	2.50	2.39	2.28	2.15	2.37	2.51	2.49	2.38	2.27	2.14	2.36
Alemow	2.35	2.30	2.21	2.05	1.79	2.14	2.30	2.25	2.16	2.01	1.75	2.09

Volkamer lemon	2.67	2.60	2.53	2.44	2.27	2.50	2.61	2.57	2.51	2.39	2.22	2.46
NRCC-3	2.56	2.49	2.40	2.24	2.07	2.35	2.52	2.50	2.36	2.20	2.04	2.32
NRCC-4	2.44	2.38	2.26	2.12	1.87	2.21	2.39	2.34	2.23	2.08	1.83	2.17
CRH-12	2.27	2.21	2.18	2.05	1.87	2.12	2.24	2.20	2.15	2.02	1.85	2.09
Mean	2.49	2.44	2.36	2.22	2.01		2.45	2.42	2.33	2.18	1.98	
C.D. at 5%	**Rootstock = 0.25 Salinity = 0.19 Rootstock x Salinity = 0.31**						**Rootstock = 0.21 Salinity = 0.15 Rootstock x Salinity = 0.30**					

Table 4. Effect of salinity on number of leaves per plant of different citrus rootstocks

Rootstocks	**Salinity level (dS m^{-1})**											
	2018-19						**2019-20**					
	0.07 (Control)	**2.5**	**4.0**	**5.5**	**7.0**	**Mean**	**0.07 (Control)**	**2.5**	**4.0**	**5.5**	**7.0**	**Mean**
Rough lemon	25.00	24.33	22.67	20.53	18.33	22.17	21.86	21.27	19.82	18.05	16.17	19.44
Cleopatra mandarin	22.67	22.33	20.87	19.27	17.33	20.49	19.69	19.39	18.13	16.74	15.15	17.82
Pectinifera	19.00	18.00	16.80	15.47	13.33	16.52	15.95	15.11	14.10	12.99	11.30	13.89
Rangpur lime	27.33	26.50	25.00	23.35	21.67	24.77	24.58	23.83	22.48	21.05	19.58	22.30
Alemow	17.33	16.33	15.17	13.87	12.53	15.05	14.27	13.45	12.49	11.46	10.40	12.41
Volkamer lemon	23.33	22.67	21.50	20.33	18.33	21.23	20.34	19.76	18.74	17.72	16.05	18.53
NRCC-3	19.33	18.50	17.67	16.33	14.00	17.17	16.33	15.63	14.93	13.80	11.92	14.52
NRCC-4	15.67	15.00	14.20	12.98	11.67	13.90	12.58	12.04	11.40	10.45	9.44	11.18
CRH-12	18.33	17.67	16.50	15.30	14.00	16.36	15.48	14.92	13.93	12.92	11.89	13.83
Mean	20.89	20.15	18.93	17.49	15.69		17.90	17.27	16.23	15.02	13.55	
C.D. at 5%	**Rootstock = 1.79 Salinity = 1.56 Rootstock x Salinity = 3.09**						**Rootstock = 1.45 Salinity = 1.23 Rootstock x Salinity = 2.57**					

Table 5. Effect of salinity on number of seedlings per seed in different citrus rootstocks

Rootstocks	Salinity level (dS m^{-1})											
	2018-19						2019-20					
	0.07 (Control)	2.5	4.0	5.5	7.0	Mean	0.07 (Control)	2.5	4.0	5.5	7.0	Mean
Rough lemon	1.58 (1.48)	1.57 (1.46)	1.54 (1.38)	1.51 (1.28)	1.43 (1.03)	1.52 (1.33)	1.57 (1.45)	1.56 (1.42)	1.53 (1.35)	1.50 (1.25)	1.43 (1.03)	1.52 (1.30)
Cleopatra mandarin	1.76 (2.08)	1.73 (2.00)	1.66 (1.75)	1.56 (1.42)	1.49 (1.23)	1.64 (1.70)	1.74 (2.04)	1.72 (1.95)	1.65 (1.72)	1.55 (1.39)	1.48 (1.20)	1.63 (1.66)
Pectinifera	1.68 (1.83)	1.68 (1.81)	1.62 (1.63)	1.56 (1.42)	1.51 (1.27)	1.61 (1.59)	1.67 (1.79)	1.67 (1.77)	1.61 (1.60)	1.55 (1.40)	1.50 (1.25)	1.60 (1.56)
Rangpur lime	1.43 (1.05)	1.43 (1.06)	1.41 (1.00)	1.41 (1.00)	1.41 (1.00)	1.42 (1.02)	1.42 (1.03)	1.43 (1.03)	1.42 (1.00)	1.41 (1.00)	1.41 (1.00)	1.42 (1.01)
Alemow	1.59 (1.52)	1.58 (1.50)	1.56 (1.42)	1.54 (1.36)	1.49 (1.22)	1.55 (1.40)	1.58 (1.49)	1.57 (1.47)	1.55 (1.39)	1.53 (1.33)	1.48 (1.20)	1.54 (1.38)
Volkamer lemon	1.60 (1.55)	1.59 (1.52)	1.57 (1.48)	1.53 (1.34)	1.48 (1.2)	1.55 (1.42)	1.59 (1.52)	1.58 (1.49)	1.57 (1.45)	1.52 (1.32)	1.48 (1.18)	1.55 (1.39)
NRCC-3	1.53 (1.34)	1.52 (1.32)	1.42 (1.00)	1.41 (1.00)	1.41 (1.00)	1.46 (1.13)	1.52 (1.31)	1.51 (1.29)	1.42 (1.00)	1.41 (1.00)	1.41 (1.00)	1.46 (1.12)
NRCC-4	1.53 (1.35)	1.52 (1.32)	1.51 (1.28)	1.48 (1.18)	1.42 (1.00)	1.49 (1.23)	1.53 (1.33)	1.51 (1.29)	1.5 (1.25)	1.47 (1.16)	1.41 (1.00)	1.48 (1.21)
CRH-12	1.62 (1.63)	1.62 (1.61)	1.56 (1.42)	1.49 (1.23)	1.41 (1.00)	1.54 (1.38)	1.61 (1.59)	1.61 (1.58)	1.55 (1.39)	1.49 (1.21)	1.41 (1.00)	1.53 (1.35)
Mean	1.59 (1.54)	1.58 (1.51)	1.54 (1.37)	1.5 (1.25)	1.45 (1.11)		1.58 (1.51)	1.57 (1.48)	1.53 (1.35)	1.49 (1.23)	1.45 (1.10)	
C.D. at 5%	**Rootstock = 0.06 Salinity = 0.05 Rootstock x Salinity = 0.13**						**Rootstock = 0.05 Salinity = 0.04 Rootstock x Salinity = 0.11**					

Original data given in parentheses were subjected to square root $\sqrt{(x+1)}$ transformation.

CONCLUSION

It can be concluded that seedling height, stem diameter, number of leaves per plant and number of seedlings emerged per seed were adversely affected when subjected to salt stress from control (0.07 dS/m) to 7 dS/m) in all rootstocks but relatively Rangpur lime, Volkamer and Cleopatra mandarin were comparatively better and Pectinifera and Alemow were poor.

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