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Full Length Research Paper

Purslane and natural vegetation as bioremediation tools to cope salinity in Satsuma mandarin orchards

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The objective of this study is to compare the Na and Cl removal capacity of natural vegetation (spontaneous flora) and cultivated purslane, as bioremediation techniques to overcome salinity in Satsuma mandarin plantations. An experimental orchard was established on two different rootstocks, (*Poncirus trifoliata* L.) Raf. and Troyer citrange (*Poncirus trifoliata* × *Citrus sinensis*)) and the orchard was irrigated at 2 different salinity levels (0.65 and 5 dSm^{-1}.The ground of the on rows of Satsuma mandarin trees was kept either with its natural flora (vegetation) or with cultivated purslane. Results are statistically examined with respect to the effects and interactions between the tested factors; rootstock, salinity and remediation tools. Biomass yields of the tested alternatives (natural vegetation and purslane) and their Na, Cl, K and Ca concentrations were measured. Also Na and Cl concentrations of mandarin leaves were analyzed to examine the remediation effect. Results showed that yield depressions of the natural vegetation plots were higher compared to purslane and showed a considerable difference according to the rootstock. Sodium removal of purslane and natural vegetation was found to be similar. Chloride uptake was lower in Troyer citrange compared to *P. trifoliata.* Purslane and/or natural vegetation can equally be recommended as Na removing bioremediation crops. Purslane can also be recommended as a promising Cl remover suitable for orchards on *P. trifoliata* (L.) Raf. rootstock.

Key words: Bioremediation, natural vegetation, purslane, salinization, satsuma mandarin.

INTRODUCTION

Large areas of arable land are being abandoned each into the roots or the vascular system (Walker and year due to misuse of soils like salinity (Essa, 2002). Douglass, 1983). The exclusion capacity can be Bioremediation of the degraded crop land is an correlated with the sterol of roots (Maas, 1993). environmentally and ecologically friendly practice. Salinity Calcium (Ca) maintains the structural integrity and causes physiological and biological disorders like osmo- selective permeability of root membranes (Hanson, tic, toxic or ionic imbalances in the plant body (Mengel 1984). The effect of salinity on nutrient composition of and Kirby, 2004). plant tissues, especially the concentration of Ca and K Chloride (Cl) and sodium (Na) are For citrus orchards, the bioremediation of the land under risky ions

*Corresponding author. E-mail: cenk.kilic@ege.edu.tr. Tel: +90 exclude Cl or Na from the scions (Maas, 1993). 232 581 63 17. Fax: + 90 232 581 71 75.

in planthas been extensively investigated (Munns, 2002; Essa, growth under excess salinity conditions. Sodium tole-2002). The authors reported that relatively high rance seems to involve the ability of xylem parenchyma concentrations of Ca and K are essential for a successful cells to extract Na from the xylem stream and sequester it growth in a saline environment. in the woody root and stem tissues (Hepaksoy et al., One cost effective friendly method to cope with salinity 1999). Chloride tolerance of plants can be heritable and is to grow salt tolerant species, varieties or to use be heritable and exclusion of Cl seems to be controlled rootstocks tolerant/resistant to saline conditions. Citrus in primarily by root membranes that either restricts Cl entry general are very sensitive to salinity. In this regard, rootstock-scion relations have been extensively studied (Aksoy et al., 2000; Hepaksoy et al., 1999). It is reported that citrus rootstocks considerably vary in their ability to salinity threat could be done by using salt removing crops either as cover crops or as intercrops between or on the tree rows or by leaving the ground with its natural flora (vegetation) to enhance salt uptake. Kumamoto et al. (1990) and Grieve and Suarez (1997) rated purslane as a very good salt removing crop according to Mass and Hoffman (1997). However, not much information is available for the salt removing capacity of natural vegetation under Mediterranean conditions. Nazik (2007), Bilen (2008) and Unal (2009) specified the yearly differences in the dominant natural vegetation species and reported that spontaneous vegetation can change yearly due to climatic conditions, main crop and fertilization. Bilen (2008) highlighted that under Mediterranean climate conditions, *Capsella bursa-pastoris* is found as the dominant flora when long term annual average precipitation was not reached. However, when the rainfall is not restricted, *Lamium purpureum* is found to dominate.

The main objective of this study was to compare Na and Cl removal capacity of natural vegetation (spontaneous flora) and the cultivated purslane (underlying intercrop) as saline bioremediation techniques for Satsuma mandarin trees budded onto two different root-stocks, *Poncirus trifoliata* and Troyer citrange. The study also aimed at determining the biomass yields, K and Ca content of these two tested alternative underlying crops. In order to make a better evaluation, Satsuma mandarin leaves were also analyzed for their Na and Cl contents with the purpose of measuring their tissue concentrations under different intercrop, rootstock and salinity conditions.

MATERIALS AND METHODS

Satsuma mandarin (*Citrus unshiu* Marc var. Owari) is a well-known *Citrus* species which ripens early in the season. An experimental plot of 0.45 ha located at Ege University Campus (Izmir/Turkey) was continuously irrigated with saline water for 13 years. The Satsuma mandarins were budded on two different rootstocks, Trifoliate orange (*P. trifoliata* (L.) Raf. and Troyer citrange (*P. trifoliata* × *Citrus sinensis*).

The plots were irrigated during the dry period (May to November) at 2 different salinity levels (0.65 dS m^{-1} (nonsaline treatment (fresh water) and 5.0 dS m^{-1} (saline treatment)) via double line source. Fresh water from a deep well was supplied through one line and highly saline stock solution was distributed through the second line. The drippers were pressure regulated and their discharge rate was 2.3 L h^{-1}. Irrigation was applied weekly based on previous day's evaporation from USWB class A evaporation pan and the crop coefficient was used as 0.5 (Doorenbos and Kassam, 1979).

Two different bioremediation tools were tested in the citrus plot, as natural vegetation and purslane. Experimental soils were sandy loam in texture with low calcium carbonate. Organic matter, N, P and K were sufficient. The EC and pH of the saturation extracts of the salinized and non salinized plots were 2.0 to 7.8 dS/m and 5.65 to 7.70 at the beginning of the study. For one season, the ground of the on rows of Satsuma mandarin trees was kept either with its natural flora (vegetation) or with purslane. In the purslane (*P. oleracea* L.) plot, the seeds were sown on a row 25 cm distant from the tree trunk, at a seeding intensity of 2 to 3 $kgha^{-1}$ (0.2 to 0.3 g m^{-2} seed). Purslane was harvested as recommended, 70 days after emergence before the formation of new seeds (Vural et al., 2000). Similarly, in the natural vegetation plot, wild species were allowed to grow spontaneously for about 120 days till the new seeds appear. The major species in the natural flora were identified by square frames (25 cm × 25 cm) which were randomly placed 5 times for each plot corresponding to a total sampled surface of 1 m^2 and their intensities (number per unit area m^2) were determined. In non salinized as well as salinized plots, number of *Urtica urens* (Stinging nettle) plants per unit area was the highest followed by *Malva sylvestris* (Mallow) and *Calendula arvensis* (Pot marigold). Generally Urtica emergence ranged from 41 to 91 per unit area.

The experimental design was split-split plot with 3 replications, the main factor being the rootstock, second salinity of irrigation water and third the bioremediation tool. Total fresh weight (biomass) of the harvested natural vegetation (g m^{-2}) and the purslane (g $plant^{-1}$) were weighed and their Na, Ca and K concentrations (% dry matter) were measured flamephotometrically and Cl potentiometrically (Kacar, 1972).

After the irrigation, Satsuma mandarin leaf samples were taken at the end of October to the beginning of November (Chapman and Pratt, 1961) and also analyzed for their Na and Cl contents according to the above stated standard methods (Kacar, 1972).

The paper presents the changes in biomass yield, Na, K, Ca and Cl concentrations of the natural vegetation and purslane and Na and Cl concentrations of Satsuma mandarin leaves. Results are statistically examined with respect to the effects and interactions between the tested variables; rootstock, salination and remediation tools as natural vegetation and purslane. All the statistical analysis was done separately for purslane and natural vegetation for tested factors. The data obtained was subjected to analysis of variance (ANOVA) and the mean differences were compared by LSD tests (Açıkgöz et al., 2004).

RESULTS

Biomass yield of purslane and natural vegetation

The main effects of salination and rootstocks on the biomass yield of purslane and natural vegetation (flora) and their interactions were statistically significant. Yield changed with respect to these factors (Table 1).

The relative yield depressions due to salination in purslane grown under Satsuma mandarin trees were close to each other, being 51% in the case of *P. trifoliata* rootstock and 57% in Troyer citrange compared to the yield obtained under non saline control conditions. Therefore, the difference between the yield depressions of purslane can be accepted as slight. On the other hand, the relative yield depressions of the natural vegetation were 78.5 and 61.6% for *P. trifoliata* and Troyer citrange rootstocks, respectively (Table 1).

Sodium, Cl, Ca and K Concentrations of purslane and natural vegetation

Sodium

Main effect of salination and rootstock on Na concentration of purslane was found to be statistically significant. On the other hand, Na concentration of the natural vegetation was affected only by salinity. The Na concentrations

Table 1. Biomass of purslane (g plant^{-1}) and natural vegetation (g m^{-2}) as a function of salinity and rootstocks.

Rootstocks	Salinity level	Fresh weight purslane (gplant^{-1})	Fresh weight natural flora (g m^{-2})
P. trifoliata	Non-salinized parcel	26.73[a]	2248[a]
	Salinized parcel	13.72[b]	1766[b]
T. citrange	Non-salinized parcel	91.28[a]	4860[a]
	Salinized parcel	52.36[b]	2994[b]
$LSD_{(0.05)}$	(Salt × Rootstocks)	3.21**	85.11**

**P<0.01. Means in the same column followed by different letters are significantly different (p≤0.05).

Table 2. Tissue Na (mg kg^{-1}) concentration of purslane and natural vegetation and as a function of salt.

Salinity level	*P. trifoliata*		T. citrange	
	Purslane	Natural flora	Purslane	Natural flora
Non-salinized parcel	957[b]	522[b]	957[a]	609[b]
Salinized parcel	1218[a]	1479[a]	2000[a]	2050[a]
$LSD_{(0.05)}$	31.01**		1185.59*	

**P<0.01, *P>0.05. Means in the same column followed by different letters are significantly different (p≤0.05).

Table 3. Tissue Cl (%) concentration of purslane and natural vegetation as a function of salinity.

Salinity level	*P. trifoliata*		T. citrange	
	Purslane	Natural flora	Purslane	Natural flora
Non-salinized parcel	0.41[a]	0.05[b]	0.47[a]	0.18[b]
Salinized parcel	0.46[a]	0.15[a]	0.67[a]	0.70[a]
$LSD_{(0.05)}$	0.072*		0.46*	

*P<0.05. Significantly different (p≤0.05).

of the two tested salt removing crops purslane and natural vegetation also differed.

The effect of salination on purslane grown under Satsuma mandarin trees on *P. trifoliata* rootstock showed higher tissue Na contents under saline conditions. Similar condition existed in the case of Na measurements of the natural vegetation. The effects of salination on purslane and natural vegetation grown under the other rootstock, Troyer citrange were determined as significant at 5% level that is higher Na concentrations were analyzed under saline conditions (Table 2). When numerically evaluated, the natural flora grown under the satsuma trees on Troyer citrange rootstock had higher Na content.

Chloride

Main effects of salinity and rootstock on Cl concentrations of purslane were found to be significant. In the case of natural vegetation, only the effect of rootstocks was statistically significant. The Cl concentrations of the two remediation tools, purslane and natural vegetation, varied according to the treatments tested in the study.

The effect of salinity showed higher Cl concentrations in both purslane and natural vegetation under both rootstock plots. The difference was significantly (5%) higher in the case of natural vegetation (Table 3).

The effects of rootstocks put forth that the Cl of purslane and natural vegetation grown under the trees on T. citrange rootstocks were respectively higher than those grown under the canopy of the trees on *P. trifoliata* rootstocks (Table 3).

Potassium

Significant effects of salinity and rootstock were deter-mined on K concentrations of purslane and of natural vegetation. Significant statistical interactions were found for the cases of *P. trifoliata* and T. citrange, salinity × remediation tools. Under salinized conditions, K con-centrations of purslane and natural vegetation decreased.

The level was almost 50% of the non-salinized conditions in case of purslane.

Table 4. Tissue K (%) contents of purslane and natural vegetation as a function of salinity.

Salinity level	*P. trifoliata*		Troyer citrange	
	Purslane	Natural flora	Purslane	Natural flora
Non-salinized parcel	1.30[a]	1.20[a]	1.47[a]	1.24[a]
Salinized parcel	0.64[b]	0.84[b]	0.77[b]	0.88[b]
$LSD_{(0.05)}$	0.118**		0.038**	

Means in the same column followed by different letters are significantly different (p≤0.05).

Table 5. Tissue Ca (%) concentration of purslane and natural vegetation as a function of salinity levels.

Salinity levels	*P. trifoliata*		T. citrange	
	Purslane	Natural flora	Purslane	Natural flora
Non-salinized parcel	0.41[a]	1.05[a]	0.43[a]	1.25[a]
Salinized parcel	0.31[a]	0.68[b]	0.38[a]	1.08[b]
$LSD_{(0.05)}$	0.168*		0.140*	

*P<0.05. Means in the same column followed by different letters are significantly different (p≤0.05).

Results related to non-salinized control conditions showed that K of the purslane under Satsuma trees budded onto *P. trifoliata* was lower than the trees where Troyer citrange was used as the rootstocks Potassium concentrations of the natural vegetation grown under trees on both rootstocks were similar when there is no salinity problem (Table 4).

Calcium

Calcium concentration of purslane was affected only by the increase in salinity levels. On the other hand, natural vegetation was significantly under the effect of both salination and rootstocks (Table 5).

The purslane and the natural vegetation under Satsuma mandarin trees with *P. trifoliata* as the root-stocks had lower Ca concentrations compared to that of those under Troyer citrange. Natural vegetation always had higher Ca than that of purslane.

Leaf Na and Cl concentrations of mandarin trees

The leaf Cl concentrations of Satsuma mandarin trees were found to be significantly higher in the salinized parcels on both of the rootstocks.

In general, similar behavior was determined for the Na contents of leaves which were also analyzed higher in the salinized trees.

Satsuma mandarin leaves had higher Cl concentrations on *P. trifoliata* than that of Troyer citrange. On Troyer citrange higher leaf Na levels were determined (Table 6).

DISCUSSION

Biomass production of purslane and natural vegetation plots under Satsuma mandarin trees budded onto *P. trifoliata* and Troyer citrange rootstocks were negatively affected by salinity. Many researchers (Maas, 1990; Kılıç et al., 2008) state similar findings supporting the results of the present study. Relative yield depressions due to salinity were found around 50% for purslane plots under these two rootstocks. On the other hand, yield depressions of the natural vegetation plots were higher and showed a considerable difference according to the rootstock.

Sodium generally does not damage the plants as does the Cl (Maas, 1993). According to the result of this study, Na removal of purslane and natural vegetation was found similar even if the rootstocks differed. It is known that Troyer citrange is a Na includer that is, excess uptake with no damage (Maas, 1990). In this case, higher Na concentrations measured in the leaves of Satsuma mandarin trees on Troyer citrange rootstock confirm this finding.

Maas (1993) and Kılıç et al. (2008) highlighted purslane as a strong Cl removing crop. Mengel and Kirby (2004) state *P. trifoliata* as Cl sensitive. The critical leaf Cl concentration for Satsuma mandarins budded on this specific rootstock is indicated to change between 0.20% (Chapman, 1968) and 0.70% (Cohen, 1976). Our laboratory results related to high Cl in the leaves of Satsuma mandarin trees on *P. trifoliata* also fall within this range. On the other hand, findings in relation to Troyer citrange showed some differences. The low Cl contents measured in the leaves of Troyer citrange can indicate its low uptake and exclusion more via roots. Consequently, it can be claimed that the residual Cl in

Table 6. Sodium and chlorine concentrations of mandarin leaves as a function of salt and rootstocks.

Salinity levels	Cl (%)		Na (mg kg^{-1})	
	P. trifoliata	T. citrange	*P. trifoliata*	T. citrange
Non-salinized parcel	0.12^{b}	0.08^{b}	386^{b}	396^{b}
Salinized parcel	0.75^{a}	0.23^{a}	728^{a}	946^{a}
$LSD_{(0.05)}$	0.053**		5.971**	

**P<0.01, *P<0.05. Means in the same column followed by different letters are significantly different (p≤0.05).

the soil is highly taken up by the natural vegetation under tree canopy. This assumption is in accordance with Cl results analyzed in the tissues of natural vegetation plots. Under saline conditions, the effect of the underlying crop kept on the orchard ground can be significant in K losses due to the nutrient competence with the satsuma mandarin trees. The tested bioremediation tools, purslane and natural vegetation, responded more or less similarly in their K removal from the rhizosphere.

The excess Ca uptake from the growing environment of citrus trees can be effectively eliminated by a suitable rootstock. The underlying plant is important as well. In the present study, natural vegetation was found suitable in removing the highest amount of Ca from the soil. Kılıç (2005) reports a small difference between Ca uptake of *P. trifoliata* and Troyer citrange rootstocks, generally the latter having higher uptake rate.

Conclusion

It is concluded that purslane and/or natural vegetation can equally be accepted as Na removing bioremediation tool(s) for Satsuma orchards established either on *P. trifoliata* or on Troyer citrange rootstocks. It is also concluded that for Cl remediation, purslane could be a suitable accumulator plant in orchards on *P. trifoliata* rootstock which is susceptible to Cl due to its high uptake affinity. Moreover, relatively lesser yield depressions in relation to Cl hazard once again showed that purslane could be suggested for *P.trifoliata*. On the other hand, under Mediterranean conditions, natural vegetation could be recommended for Satsuma mandarin trees budded on to Troyer citrange rootstock. Troyer citrange rootstock excludes the excess Cl via its roots.

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