ROOT ANATOMICAL PLASTICITY IN RESPONSE TO SALT STRESS UNDER REAL AND FULL-SEASON FIELD CONDITIONS AND DETERMINATION OF NEW ANATOMIC SELECTION CHARACTERS FOR BREEDING SALT-RESISTANT RICE (*Oryza sativa* L.)

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Abstract: Specific understanding of root anatomy plasticity under salt stress is lacking and requires creation of efficient screening techniques for stress condition s. To fill this gap, this study aimed to determine the anatomical plasticity in root chracteristics of 31 different rice cultivars (from 'Best' to 'Low' yielding) grown under real field conditions (saline and non-saline) from planting to harvesting and to reveal detailed root anatomical parameters that can be used to select and breed salt-tolerant rice. Anatomical and histochemical features of all cultivars and thin structures of the apoplastic barriers were investigated. The amount of silica (Si), 35 different anatomical characteristics, anatomical plasticity characteristics, plasticity rates, plasticity trends and changes and strategies of each group under saline and non-saline conditions were compared. The results showed that protective anatomical characters improved/remained equal to, and worsened/remained equal to those of the controls, in the 'Best' and other groups, respectively, from non-saline to saline conditions. Anatomical plasticity is essentially directly related to apoplastic barrier features. High genotypic variation was observed in root anatomy in all cultivars, but foremost traits were as follows: (1) cell size, (2) Si presence, (3) Si accumulation shape, (4) Si distribution towards root stele, (5) xylem arch features, (6) lignification-suberization properties in apoplastic barriers and their degrees, (7) presence/absence of idioblast cells filled with gummic and phenolic substances and (8) moderate anatomical plasticity. Cultivars with the most stabile anatomy under saline and non-saline conditions should be used to select and breed salt-resistant rice.

Key words: Oryza, rice, salt stress, anatomy, breeding, selection

Gerçek ve Tam Zamanlı Tuz Stresi Koşullarında Çeltikde (*Oryza sativa* L.) Değişken Kök Anatomisi Özellikleri ve Tuza Dayanıklı Çeltik Islahına Dönük Yeni Anatomik Seleksiyon Karakterlerinin Tespiti

Özet: Kök anatomisinin tuz şartlarında kullanılabilirliğine dair bilgiler eksiktir ve bu anlamda etkin anatomik seleksiyon karakterlerinin tespit edilmesi, çalışmanın esas amacını oluşturmaktadır. Bunun için 31 değişik çeltik çeşidinin (verim değerlerine göre "En iyi"den "en düşük" ekadar) tuz şartları ve kontrol şartlarında, çimlenmeden hasada kadar, karşılıklı olarak kök anatomik özellikleri karşılaştırılmış ve detaylı anatomik özellikleri çıkarılmıştır. İlaveten histokimyasal yöntemlerle apoplastik yapılar araştırılmıştır. Si elementi değerleri, anatomik değişken değerleri, plastisite oranları, plastisite eğilimleri ve her grubun tuz stresi altındaki stratejik anatomik değişim yönelimleri tespit edilmiştir. Sonuçta; koruyucu anatomik karakterlerin "En iyi" grupta ya aynı kaldığı (kontroldeki gibi) ya da kısmen iyi yönde artış gösterdiği, diğer gruplarda ise aynı kaldığı (kontroldeki gibi) ya da daha kötü yönde değişim gösterdiği tespit edilmiştir. Anatomik plastisite özelliğinin temelde apoplastik bariyer karakterleri ile doğrudan ilişkili olduğu ortaya çıkarılmıştır. Her ne kadar tüm çeşitlerde tuz stresi altında az ya da çok anatomik değişkenlik görülse de en önde gelen anatomik karakterler şunlardır: (1) hücre boyutları, (2) Si varlığı, (3) Si birikim şekli, (4) Si'un kök stelar kısma doğru dağılımı, (5) ksilem ark yapısı, (6) apoplastik bariyerlerin lignifikasyon-suberinizasyon özellikleri ve dereceleri, (7) zamk ve fenolik içerikli idioblastik hücrelerin varlığı/yokluğu, (8) orta derecede anatomik değişkenlik. Özetle; tuzlu ve normal şartlar altında koruyucu modifikasyonlarını en stabil tutan çeşitlerin, seleksiyon ve ıslah açısından en fazla dikkate değer olduğu tespit edilmiştir.

Anahtar kelimeler: Oryza, pirinç, tuz stresi, anatomi, ıslah, seleksiyon

Introduction

Rice (*Oryza sativa* L.) is the most salt-sensitive crop because it is ineffective in controlling the influx of salt (Na⁺) into the roots, causing rapid salt accumulation at toxic concentrations in the plant (Singh & Flowers 2011). The effects of salinity on rice include reduced seed germination

(Hakim et al. 2010), decreased growth and survival of seedlings, damage to the structure of chloroplasts (Yamane et al. 2008), reduced photosynthesis (Moradi & Ismail 2007) and decreased seed set and grain yield (Asch et al. 2000). To cope with salinity, salt-tolerant rice cultivars are

needed to be developed (Rajendran et al. 2009), which requires the development of efficient techniques for identifying the components related to salt tolerance (Ashraf & Akram 2009). When rice plants are exposed to salt stress, they generally respond to this stress at morphological, anatomical, cellular and molecular levels (Ashraf 2004). On the other hand, some studies have reported that salt resistance can be achieved by physiological factors without considering anatomical parameters (Hwang & Chen 1995). However, in a previous study evaluating factors involved in overcoming drought stress in rice, effective root anatomical parameters were revealed and substantial genotypic variations in the root system were observed, which can be used to enhance the water capture ability and improve the drought tolerance of rice (Fukai & Cooper 1995, Meyer et al. 2009). In addition, structural variations were observed when roots were treated with different drugs (Fang et al. 2007, Suralta & Yamauchi 2008, Meyer et al. 2009, Lynch et al. 2014). These findings observed under different stress conditions suggested that root anatomical variations under saline conditions could be used as a great opportunity for salt-resistant rice breeding. The lack of efficient root anatomical screening techniques has delayed progress research on salinity resistance in rice (Singh et al. 2013, Lynch et al. 2014).

On the other hand, because of the complexity of interactions environmental such as soil-water interactions, real and full-season studies that integrate in vivo approaches, such as root function and/or anatomy assays, will be important tools in concert with empirical studies. This change will require expertise in plant biology and breeding studies. Most importantly, this challenge calls for renewed emphasis on understanding the plant root phenomene in the context of salt stress (Lynch et al. 2014). In summary, there has been a lack of studies detailing rice root anatomical parameters across many cultivars under full-season and real salt-stress conditions. In addition, more effective and practical root anatomical parameters involved in the response of rice plants to salt stress have not been determined. In this study, we examined rice root anatomy under real saltstress environmental conditions and specifically noted which parameters exhibited changes, if any, and to what degree could they be used in screening for salinity resistance. In summary, this study had the following aims: (1) to examine root anatomical parameters of different rice cultivars which were categorized into 4 different groups (Best, Good, Middle and Low) according to their yields and salt tolerance under saline and non-saline (control) conditions; (2) to determine the extent of anatomical modifications (plasticity) of 31 different rice cultivars grown under saline and non-saline real field conditions and in all soil-water interactions from germination to harvest; (3) to measure strategic responses of root anatomy under natural salt-stress conditions and (4) to develop an effective screening procedure for rice resistance to salinity based on the results.

Materials and Methods

All experiments were conducted simultaneously in Edirne, TURKEY, in Thrace Agricultural Research Institute's fields (control) along Meric River and in a field with salty water and soil conditions in Salarlı village along Ergene River, Uzunköprü (Fig. 1, 2). The average temperature data of the fields' area and all water + soil analysis averages were obtained from official meteorological state and regional agricultural research institute routine laboratory, respectively. The day temperature ranged from 25°C to 34°C, while the night temperature fluctuated between 22°C and 31°C. The humidity ranged from 50% to 75% during the growing period. Chemical characteristics of the soil and water were as follows; Ergene basin, saline conditions; soil pH: 7.47 (light alkali); 1.591.00 mmhos/cm; water pH: 7.96; ECx108: 3580 micromhos/cm; sodium absorption ratio (SAR): 18.71; and irrigation water class: C4S3 (very high salt concentration, not suitable for irrigation). Meric non-saline conditions; soil pH: 7.05 (neutral); water pH: 7.49; ECx108: 630 micromhos/cm; SAR: 5.64; and irrigation water class: C2S1 (a good level of irrigation water, can be used as irrigation water in almost all plants).

Thirty-one different rice cultivars with varying salt resistance (categorised as Best, Good, Middle and Low) and yield (Aybeke & Demiral 2012) were used in this study (Table 1). Roots were washed with the same river water after removing the soil. The silica (Si) concentration in the roots was determined following nitric-perchloric acid digestion using inductively coupled plasma optical emission spectrometry (ICP atomic emission spectrometer, Perkin-Elmer Co., Norwalk, CT, USA) (see also Aybeke & Demiral 2012) and Si content was represented by its concentration (% weight) in the roots. In all trials, 85-day and 14-leafed mature flowering plants were used (Sürek 2002). In the field, upper one third parts of adventitious roots were cut by using lancet and fixed in formalin-acetic acid-alcohol mixture (Aybeke 2004). These samples were washed and stored with 96% and 70% alcohol, respectively. Their paraffin sections were made with a Leica RM2255 microtome and three different staining methods were applied (Hematoxylin-Eosin, Alcian Blue-Safranin and Sartur staining for ergastic substances). The sections were incubated for 3-5 minutes in hematoxylin and washed with tap water before stained with eosin for 10-15 seconds. For Safranin-Alcian Blue method, paraffin sections were treated with 6 parts Safranin-4 parts Alcian Blue dye mixture for 3 minutes and washed with rising alcohol serie. For Sartur dye method (Celebioğlu & Baytop 1949) sections were directly examined by using 1-3 drops of the dye under a microscope for ergastic substance detection. All dyed sections, except Sartur, were mounted on glass slides with Entellan after passing the xylene series. Microphotographs were taken with an Olympus BH-2 photomicroscope and anatomical investigations were made under an Olympus BH-2 photomicroscope. Thirty-five different qualitative anatomical parameters of the roots were determined (Table 1). Anatomical plasticity trends of each cultivar were calculated by collection of differences Root Anatomical Plasticity in Response to Salt Stress Under Real and Full-Season Field Conditions and New Efficient Screening Techniques for Breeding Salt-Resistant Rice (Oryza sativa L.)

(improved/ worsened/remained unchanged, as symbols \blacktriangle , \blacktriangledown , # in Table) in anatomical traits across treatments. This characterized the strategies of each group under salt stress. Anatomical change rates of each group were found by comparing anatomical changes under stress conditions in different root tissues. No statistical analysis was performed because all parameters were qualitative (not quantitative). In addition, root apoplastic barrier properties and Si content of some cultivars were investigated by EDX facilitated scanning electron microscopy (SEM). For SEM preparation, the method of Aybeke (2007) was used with some modifications. Root sections were directly observed and photographed with a Zeiss EVO LS10 scanning electron microscope after paraffin dissolution.

ABBREVIATIONS: A: Arches, C: Cortex, E: Exodermis, ED: Endodermis, EI: Exodermis inner, EM: Exodermis middle, EO: Exodermis outer, IC: Inner cortex, M: Medulla, OC: Outer cortex, P: Pericycle, PH: Phloem, Prt: Protoxylem, Uth: 'U' thickening, Xy: Xylem.

Results

Epidermis was generally quitely damaged, mostly lost, 3-layered exodermis (outer layer, lignified middle layer, partially or non-lignified inner layer), outer cortex (OC) present or not, as a continuous or alternating with Exodermis inner (EI), aeranchymatic middle cortex, 1-2layered, Inner cortex (IC) sometimes absent, non-lignified or slightly and partially lignified, and thin-walled, not usually "U" form of thickening (Uth) in the Endodermis (ED), if it exist, thin; Pericycle (P) as a continuous line, Phloem (PH) usually in the form of small island between arches, parenchyma intact, robust, protoxylem and arces were generally regular, Arches (A) 3-4, Medulla (M) thin, slightly thick-walled and lignified (Fig. 3a).

General anatomical features of the groups

Exodermis;

Best group: EO: 1 row; EM: regular, quite thick-walled; EI: 1 row and large (Fig.3b).

Good group: EO: 1 row, more or less damaged; EM: 1–2-layered, in which all walls lignified; EI: 1 row, thinwalled or relatively thick-walled, lignified to different degrees.

Middle group: EO: 1 row, regular, sometimes largecelled; EM: 1–2-layered in rows; EI: 1 row, lignified in various thicknesses.

Low group: EO: 1 row, rarely 2-rowed or damaged (Fig. 3c); EM: 1-2 rows, with walls varying in thickness; EI: 1-2 rows, absent or alternating with OC, with or without lignification.

Cortex and endodermis;

Best group: OC: alternating with E or absent, aerenchyma lysigenous, less lignified or non-lignified; IC: 1–2 rows, partly lignified or non-lignified; ED: 'Uth' is thin or relatively thick.



Figure 1. The satellite map of the areas where the field trials were performed (the maps were obtained from Google Earth program. **a**) Control (non-saline) conditions (Meriç River), **b**) Saline conditions (Ergene River). Red stars indicate the experimental fields and yellow arrows indicate the rivers.

Good group: OC: absent or partly present, aerenchyma usually lysigenous and containing some amounts of Si; IC: absent or partially present, thin-walled; ED: absent or 1 row; 'Uth': unclear or obvious, generally thin-walled.

Middle group: OC: absent or present, aerenchyma lysigenous and sometimes lignified; IC: 1–2 rows, regular, lignified or non-lignified; ED: 'Uth' indistinct or clear.

Low group: OC: absent or alternating with IE, regular and crushed, aerenchyma lysigenous or partially schizogenous, lignified or non-lignified, rarely with Si; IC: 1–2 rows, damaged, regular, thin-walled, lignification variable; ED: 'Uth' irregular, very fine (Fig. 3d) and damaged.



Figure 2. Field view of 31 rice cultivars grown in saline Ergene conditions.

In Meriç conditions:

Table 1. Rice cultivars of which root anatomical qualitative parameters examined in the study. Abbreviations written in red and *italics* are additional characters specific to anatomical plasticity table and others (black) are characters that are common to anatomical properties and anatomical plasticity (changing) tables. *: yield values of each cultivar and categorizations of all of them were carried out according to Aybeke & Demiral (2012).

Cultivars and grouping in terms of yielding values*	 Best: Kral, Kırkpınar Good: 7721, Sürek, Ece, Kros 424, Gala, Veneria Middle: Altınyazı, Durağan, Halilbey, Koral, N-41-T, Osmancık Low /very low: Akçeltik, Beşer, Edirne, Gönen, İpsala, Karadeniz, Kargı, Meriç, Neğiş, Plovdiv, Ranbelli, Rocca, Şumnu, Trakya, Tunca, Yavuz, Kızıltan
Exodermis	 EOL: Outer Exodermis layer number G: Greatness, G: big, GG: layer number increased to 2, and both two layers robust, G-G: the outer deformed, inner one is most stable, g: normal-small S: structure, +: regular, ++:uniformity more increased, -: damaged, ±: partially damaged/regular AW: Anticlinalwall, II: ant. walls erect, I<: erect-lobed; <: lobed S: silica, s+: present, s++: silica has increased even more so when, s-: absent, S+*: silica on outer periclinal walls, s+**: silica on the corners of cells EML: Middle Exodermis layer number W: wall, w: thin, w-: wall thin and more thinned under salinity, W: thick, WW: fairly thick, w*: silica on the corners S: structure, +: regular, -: damaged, ±: partially damaged / regular, +,si: both regular and bearing silica EIL: Inner Exodermis layer number, 0: absent/ damaged, 1,2: present, *: cells large, **: become larger, s: become smaller, ↔: alternating with inner cortex L: lignified wall, -:absent, □: on all surface, ∩: on only outer periclinal and anticlinal walls, ∩ ▲: lignified iterly W: wall, +: thick-walled, □: on all surface thick, ∩: outer periclinal walls and anticlinal walls thick, -: not thick(thin-walled), Ss: with silica, ↔: alternating with outer cortex
Cortex/Endodermis	 12. OC: Outer cortex, layer number (1,2), -: absent, ↔: alternating, wintower cortex 13. AE: Aeranchyma, lz: lizigen, sl: schisolysigenous 14. L: lignification, l+: present, l-: absent, l±: partially found, ∩: outer periclinal walls and anticlinal walls lignified, □: on all wall surface lignified 15. Si: silica, s+:present, s-:absent, 16. IC: Inner cortex layer number (1,2), 0:absent 17. L:lignification, l+: present, l-:absent, l±:partially present, ∩: only outer periclinall walls, □: on all wall surface 18. W: wall, w: thin-walled, W: thick-walled, WW: more thick-walled, w+s: both thin-walled and with silica, ∩: only outer periclinal walls thick, □: all wall surface thick, 19. R: Regularity, +: regular, -: damaged, ±: regular-damaged in from places to place, ++: while regular control, become more regular salt stress 20. E: endodermis, 0: absent, 1: 1-layered, 21. R: Regularity, +: regular, -: damaged, ±: regular-damaged in from places to place, 22. U: thickening type (Figure 16-22)
Pericycle, Phloem	 22. U: thickening type (Figure 16-22) 23. PS: pericycle structure, p+: regular, p-: unregular, p++: while regular in control, but more regular in salt stress 24. P#: pericycle layout, f: pericycle dotted with phloem, p: percycle continuous, not dotted, f: pericycle dotted the same manner as in non-saline conditions, f-: interruption reduced, ff: interruption increased, L+: lignified, W: thick-walled, WW: more thick-walled 25. PH: phloem, fp↑: phloem located both on pericycle and in stele, f=: phloem only on periciyle 26. PP: gap status in pericyle, p0: pericycle includes gaps, p1: pericyle is as an continuous layer, not gapped 27. Si: silica, -: absent, 1: present, ±: present partly, id: idiyoblast cells present 28. PH: phloem ve parenchyma layout, altogether, PH+: regular, PH-: unregular, PH±: damaged place to place, PH++: while regular, became more regular in salt stress, L+: additionnaly lignified under saline stress, W: thick-walled in saline conditions

ble 1. Continued	29. A: arches number, $\mathbf{\nabla}$: decline, $\mathbf{\Delta}$: increased, = not changed							
	30. AS: arches structures, a +: regular, a -: unregular, a ±: partly damaged / regular, w : thin-walled, * lignification occurred a little more than usual, <i>W</i> : thick-walled, L±: Lignified more or less, not quite							
	31. M: medulla, w: thin-walled, W: thick-walled, WW: fairly thick-walled, WWW: extremely thick walled, W±, w±: wall has been damaged in salt stress							
Xylem	32. Si: silica, +: present, -: absent, ±: present place to place							
	33. S: Stelar layout, L ▲: lignification increased, L±: more or less lignified, W ▲: thickness increased -: unchanged, id: idiyoblast cells present, S ▲: stele became more regular and robust							
	34. Pr: protoxylem, -: absent, / little, 1: at normal density, 2: very dense, ▲: density increaesed							
	35. Prl: protoxylem lignification, pr+: normal, lignified, pr±: partly lignified, pr-: non-lignified, L + lignification increased, W: thick-walled, A: integrated with arches							

Pericycle and stele;

Best group: P: interrupted with PH (also in other groups); PH and Xy: parenchyma regular; M: normal or quite thick-walled, regular (Fig. 4a).

Good group: P: quite crushed; PH: located in both P and stele, Si present sometimes, generally PH and parenchyma damaged or regular; A: very damaged or regular; M: thin or very thick-walled, lignified, included Si in some samples (Fig. 4b). Middle group: PH and parenchyma regular or irregular, crushed, thin-walled; A: 4–5, arcs with Si partially available (Fig. 4c), protoxylem lignified.

Low group: P: irregular, damaged or relatively regular; PH and parenchyma: regular or irregular; A: 3–7, trachea partially or completely damaged, rarely with Si; M: thickness and lignification variable, sometimes include Si. In addition, all anatomical features of all cultivars under non-saline conditions (Meriç) are given in Table 2 in detail.

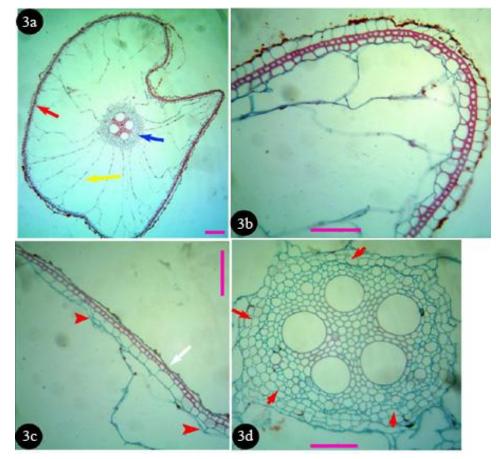


Figure 3. a) General anatomical features of rice root. Red, yellow and blue arrows indicate exodermis, aerenchyma and stele, respectively. b) Kırkpınar (Best) root exodermis in control conditions. All three exodermal layers are regular. The middle layer is small, clearly lignified and thick-walled, c) Ranbelli (Low) in control conditions. The outer exodermis is almost completely lost or fairly flattened (arrow) and the outer cortex is entirely absent or small-celled (arrow head), d) Ranbelli (Low) in control conditions. Low endodermal U thickness (arrow) is seen. **Bars:** 50 μ in **3a** and 20 μ in **3b-d**.

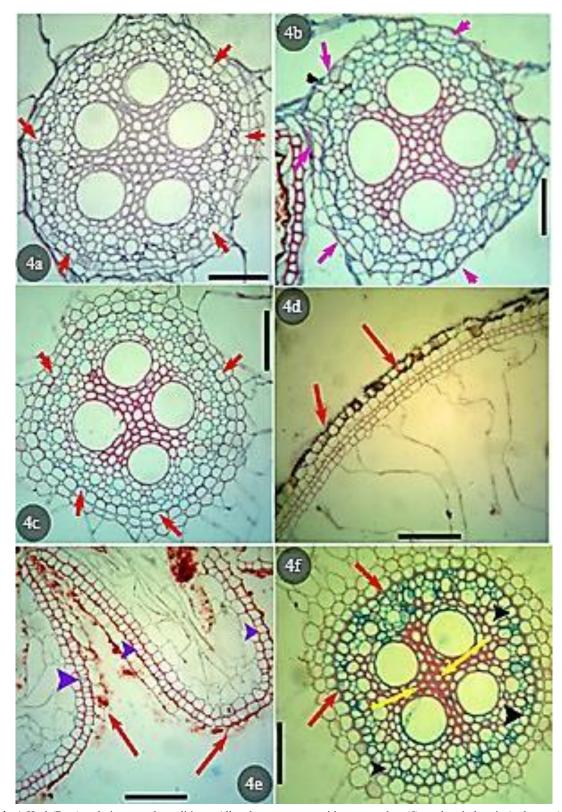


Figure 4. a) Kral (Best) stele in control conditions. All stelar structures, with more arches (5), and endodermis (red arrow) are very regular, **b**) Kros (Good) stele in control conditions. A partially crushed inner cortex (arrow) and irregular endodermis (arrowhead) are seen, **c**) N41T OT (Middle) stele in control conditions. There is a regular and continuous endodermis (arrow) and all standard stellar structures (**a**: arches, **m**: medulla), **d**) Kırkpınar (Best) exodermis in Ergene conditions. Note the clear and regular exodermal cell layers, particularly the outer cells densely filled up with suberized material (red arrow), **e**) Ece (Good) exodermis in salty Ergene conditions. The outer layer is partially damaged (arrow), and there is hin-walled middle layer (arrowhead), **f**) Kargı (Low) in salty Ergene conditions. The endodermis "U" structure (red arrow) and medullar cells (yellow arrow) are very thick and the pericycle (arrowhead) is lignified. **Bars:** 20μ .

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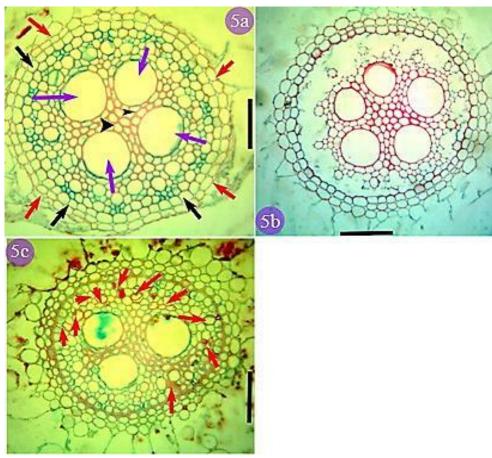


Figure 5. a) Kral (Best) stele in salty Ergene conditions. Very clear and regular inner cortex (red arrows) as well as endodermis (black arrows) arches (blue arrows) and medullar cells (arrowhead) were seen, b) 7721 (Good) stele in salty Ergene conditions. The phloem and xylem parenchyma were fairly damaged and were lost, c) Yavuz (Low) in salty Ergene conditions. Dense stellar lignification was observed along with idioblast cells filled with gummic and tannic/phenolic substances (arrows). **Bars:** 20µ.

In salty Ergene conditions:

Exodermis;

Best group: EO: 1–2, slightly damaged, regular, sometimes with Si; EM: small, lignified, 1–2 rows, thin-walled (Fig. 4d); EI: alternating with OC or continuous.

Good group: OE: 1–2 layered, Si intense; EM: 1 row, rarely 2 rows, thin-or thick-walled, regular (Fig. 4e); EI: absent or present, 1 row, alternating with OC, small or large-celled, lignified.

Middle group: OE: 1–2 rows, large, crushed, with more or less dense Si; ME: 1–2 rows, lignified and continuous, sometimes with Si; EI: absent or alternating with cortex, lignified in different ways.

Low group: OE: absent or 1–2 rows, sometimes with Si, large, sometimes crushed; ME: 1-layered, walls lignified, small, regular, with Si present sometimes; EI: 1 row, absent or replaced with OC, sometimes locally damaged by Si.

Cortex and endodermis;

Best group: aerenchyma lysigenous (common in almost all groups); IC: 1–2 rows, regular, lignified; ED: 'Uth' very thin or too thick.

Good group: aerenchyma non-lignified, with dense Si, non-lignified; IC: 1–2rows, lignified; ED: 'Uth' at different rates.

Middle group: OC: sometimes absent or regular, large or alternating with EI, aerenchyma sometimes schizogenous, partially lignified; IC: 1–2rows, rounded, lignified, and thick-walled; ED: 'Uth' thin or regular.

Low group: OC: absent or alternating with EI, aerenchyma partially schizo-lysigenous, non-lignified or lignified; IC: absent or 1–2 rows, regular, lignified, partly thick-walled; ED: 'Uth' uncertain or partly thick (Fig. 4f).

Pericycle and stele;

Best group: P: interrupted with PH (almost common in other groups); PH: located in both P and stele (common); PH and parenchyma: thin-walled; A: 4–6; M: thick-walled; Xy: regular; protoxylem at normal density, thick-walled, lignified (Fig. 5a).

Good group: P: 1 row, sometimes lignified, idioblast cells, Si and lignified cells were found; PH: regular, rarely damaged (Fig. 5b); A: 4–5; M: thin or thick.

Middle group: P: rarely continuous (uninterrupted), idioblast and dense lignified cells present; parenchyma:

Low group: P: sometimes lignified and thick-walled, PH, parenchyma and stele regular or partly damaged; A: 3-7, trachea/tracheid sometimes damaged or lignified intensively, sometimes stele not fully lignified or excessively lignified, tannic substances, idioblast cells present (Fig. 5c); M: variable in thickness. Anatomical properties or anatomical plasticities of all cultivars under saline conditions (Ergene) were given in Table 3 in detail. In general, many of the characters in the Best group remained unchanged. Also changing trends which is positive and negative direction, almost equal to each other; as for other groups (Good, Middle, Low), changing trends of the characters became more active, reduced the number of unchanging character. Another important finding is that when switching to the normal conditions to saline conditions, anatomical characters vary more positively, especially Good, Middle and Low groups.

Strategic approaches of groups for removing salt stress

Exodermis;

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Best group: Strategic parameters (Table 4) were more stable in this group. Also, E-C alternation was absent, which was an important difference from the other cultivars. If any of these parameters were missing, the salt stress would be overwhelming. The modifications of both EO and EI layers were as important as those of the EM.

Good group: EO generally exhibited more uniform changes, was large and had more rows of cells and Si. EM row number generally decreased from control to salty conditions. Walls were transformed partially or entirely to being thin lignified or unchanged. EI showed increasing or decreasing modifications in cell size or lignification or both. Within this group, inverse changes among these strategic parameters (Table 4) were frequent. In general, in the groups 'Best' and 'Good', protective variations were reduced in EM, but concentrated in EO and EI.

Middle group: Osmancık, Durağan and Koral exhibited more protective variation than other cultivars in this group. EM usually exhibited a thinner and more lignified wall, the row number remained unchanged or decreased and Si was present. EI, particularly in Koral and Osmancık, was superior in terms of the cell size, robustness and lignification.

Low group: Akçeltik, Beşer and Edirne showed more considerable protective mechanisms than others, but Kargı, Plovdiv and Yavuz were very different from the others in terms of the cellular layout. As results; in this group, increasing modifications in outer and inner layers were partially seen as to be changed from lower level to normal level.

Cortex and endodermis;

Best group: Kral kept its same feature of Meriç and showed no change under salt stress conditions. In contrast,

Kırkpınar exhibited fundamental changes in ED as well as IC. No change was found in OC, and variations were mainly found in IC and ED.

Good group: EO and EI of 7721, Kros-424 and Gala also exhibited considerable modifications. In contrast, variation in IC increased inversely with that in OE for Ece.

Middle group: OC was more apparent, regular, large or alternated with EI. Modifications in C-E were not as substantial as those in EO. In contrast, these modifications shifted to IC and/or OC and ED.

Low group: All results are given in Table 5 as well as an indication as to whether Si was elevated or not.

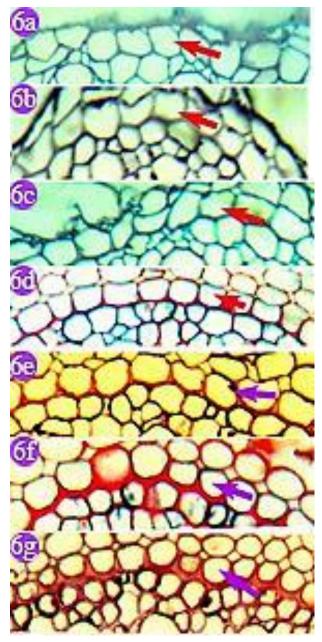


Figure 6. Endodermis types according to their 'U' wall thickness; Meric types: **a**) Tip 0, **b**) Tip I, **c**) Tip II. Ergene Types: **d**) Tip I, **e**. Tip II, **f**) Tip III, **g**) Tip IV.

Root Anatomical Plasticity in Response to Salt Stress Under Real and Full-Season Field Conditions and New Efficient Screening Techniques for Breeding Salt-Resistant Rice (Oryza sativa L.)

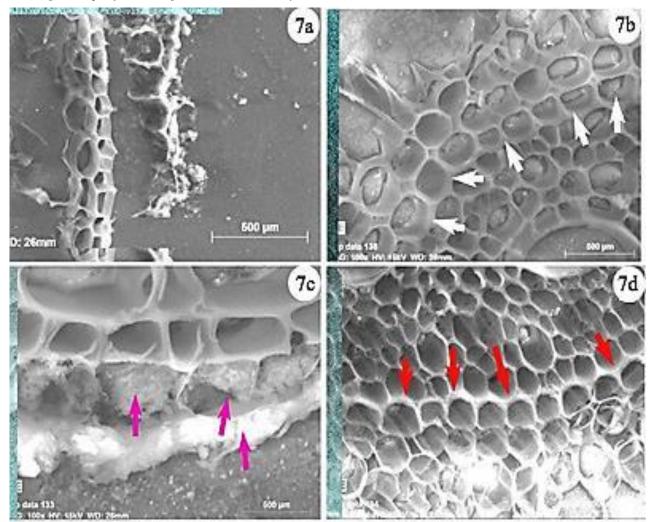


Figure 7. a) Partially magnified exodermis of Akçeltik root (outer layer damaged thoroughly, b) Akçeltik endodermis (arrows), c) Kırkpınar (salty Ergene conditions), suberised, thick-walled outer exodermal cells, covered externally with dense suberin and filled with dense suberised material (arrows), d) Endodermis (EDX densities were schematized on the same figures as sidebars indicating Si contents as blue points densities from outward to inward).

Pericycle and phloem;

Best group: With regard to P continuity, Kral was better than most cultivars.

Good group: Usually PH was cut intermittently by P (common), but variations in discontinuity occurred.

Middle group: Gaps in P were either present or absent. N-41-T, Durağan and Osmancık showed more protective variations than the other cultivars. PH and parenchyma layouts did not worse nor did they remain unchanged in the Best, Good and Middle groups. In the Low group, these features generally worsened.

Xylem;

Best group: There was no reduction or worsening change in Kırkpınar.

Good group: In particular, Sürek and 7721 showed increased protective modifications in ED, C, P and Xy.

Middle group: In particular, Xy of Osmancık changed significantly.

Low group: Idioblast cells were observed in Tunca and stellar lignification increased considerably in Yavuz, Rocca, İpsala and Akçeltik (Table 4).

The plasticity rates had a remarkable increase depending on different root regions (Table 5).

In general, under saline conditions, the Si content of all cultivars increased, whereas cultivars in the Best group showed little change or a slight increase in Si content. According to the Si value in all groups, Si content was not a distinguishing character (Table 6). Under saline conditions, in Akçeltik (Low group), Si contents [as total amount (%weight)] were 414776 (68.55%) and 358071 (68.59%) in E and ED, respectively, according to EDX-SEM examination (Fig. 7a-7b). As for Kırkpınar, Si amounted to 344177 (68.30%) in E and 333611 (62.92%) in ED. Furthermore, the outer exodermal cells are densely filled with suberin material as well as Si. (Fig. 7c-7d).

Groups*		Outer	Exo	lerm	is	Mid	ldle Ex	od.	Inne	r Ex	od.	Outer	cortex	, Aeran	chyma	In	ner	corte	ex	End	loder	mis		Peri	cycle	/ Phl	loem				Xyle	m		
	EOL	G	S	AW	s+/-	EML	W	S	EIL	L	W	OC	AE	L	Si	IC	L	W	R	E	R	\mathbf{U}^2	PS	P#	PH	PP	si	PH	Α	AS	Μ	Si	Pr	Prl
Kral	1	G	+	_I<	s+*	1,2	WW	+	1,*↔	-	-	\leftrightarrow	lz	Ŀ	S-	1	Π	W	+	1	+	Ι	P+	f	fp↑	P1	-	PH+	5	a±	W	-	1	Pr+
Kırkpınar	1	G	+	<	s+*	1,2	WW	+	1*		-	0	1z	1-	S-	1,2	Π	w	+	1	+	I,II	P+	f	fp↑	P1	-	PH±	5	a+	WW	-	2	Pr+
7721	1	g	-	<	S-	1,2	W	+	1*	Π	-	-	lz	1-	S-	0	-	-	-	0	-	0	P-	f	fp↑	P1	-	PH-	-	-	-	- 1	-	-
Sürek	1	Ğ	+	I<	s+*	1,2	W	+	1*		Π	-	1z	1-	S-	1	1-	w	-	1	+	0	P+	f	fp†	P 1	-	PH-	4	a-	W	- 1	1	Pr+
Ece	1	G	±	I<	S-	1,2	WW	+	1		+	0	sl	l±	S-	1	\cap	w	+	1	+	II	P+	f	fp↑	P 1	-	PH±	6	a+	WW	- 1	1	Pr+
Kors-424	1	g	-	<	S-	1	W	+	0	-	-	0, ↔	lz	1-	S-	0	-	-	-	1	-	Ι	P-	f	fp↑	P1	-	PH-	4	a+	W	+	1	Pr+
Gala	1	g	-	<	s+	1,2	W	+	1, ↔	Π	-	0	lz	l±	s+	1	1-	w+s	+	1	+	Ι	P+	f	fp↑	P 1	±	PH+	4	a+	W	- 1	1	Pr±
Veneria	1	g	±	I<	s+	1,2	WW	+	1		-	\leftrightarrow	lz	L+	S-	1	l +	W	-	1	+	1	P+	f	fp↑	P1	-	PH+	4	a+	WW	-	2	Pr+
Altınyazı	1	g	±	II	s+	1	WW	+	1	\cap	-	-	lz	L+	S-	1	l +	W	+	1	+	0	P+	f	fp↑	P1	-	PH+		a±	W	±	1	Pr+
Durağan	1	G	±	<	S-	1	W	+	1		-	$1, \leftrightarrow$	lz	1-	S-	1	1-	W	-	1	-	0	P+	f	fp↑	P1	-	PH±	4	a+	W	-	1	Pr+
Halilbey	1	g	-	<	s+*	2	WW	+	1	\cap		0	lz	1-	S-	1,2	1-	W	+	1	+	0	P+	f	fp↑	P0	-	PH-	5	a±	W	-	2	Pr+
Koral	1	G	-	<	S-	1	WW	+	0	-	-	-	lz	1-	S-	1	Π	W	+	1	+	0	P+	f	fp↑	P1	-	PH±	5	a+	W	-	2	Pr+
N-41-T	1	g	-	I<	S-	1,2	W	+	1		\cap	0	1z	l±	S-	1,2		W	+	1	+	Ι	P-	f	fp↑	P1	-	PH+	4	a+	WW	- 1	1	Pr+
Osmancık	1	g	±	<	S-	1,2	W	+	1*	\cap	-	0	lz	l+	S-	1	\cap	W	-	1	-	0	P+	f	fp↑	P1	-	PH-	5	a-	W	-	-,1	Pr±
Akçeltik	-	-	-	-	-	1	W	+	0	-	-	0	lz	l±	S-	0,1	1+	W	-	1	-	0	p-	f	fp↑	P1	-	PH-	5	a±	W	+	1	Pr+
Beşer	1	g	+	I<	s+	1,2	WW	+	1	\cap	+	-	lz	l±	S-	0,1	l±	W	±	1	+	Ι	P+	f	fp↑	P1	-	PH±	5	a±	W	±	1	Pr+
Edirne	2	G	+	I<	s+	1,2	WW	+	$1, \leftrightarrow$	\cap	\cap	\leftrightarrow	lz	1-	S-	1,2	Π	W	+	1	±	Ι	P+	f	fp↑	P1	-	PH+	7	a±	WW	-	2	Pr+
Gönen	1	G	+	I<	S-	1	WW	+	1,2	-		1	lz	1-	S-	1,2	1-	W	+	1	+	0	P+	f	fp↑	P1	-	PH+	5	a+	WW	- 1	1	Pr+
Ípsala	1	G	±	I<	S-	1,2	WW	+	1	Π	-	\leftrightarrow	sl	1-	S-	1	1-	W	+	1	+	0	P+	f	fp↑	P1	-	PH+	5	a±	W	- 1	2	Pr+
Karadeniz	1	G	±	<	s+*	1	WW	+	1	Ω	Π	0	lz	l±	S-	1	l±	W	+	1	+	0	P+	f	fp↑	P1		PH+	4	a+	WW	+	1	Pr+
Kargı	1	G	-	<	S-	1	WW	+	1*	Π	-	0	lz	1-	S-	1	Π	W	+	1	+	I,II	P+	f	fp↑	P1	-	PH+	3	a+	W	-	1	Pr+
Meriç	1	g	±	<	s+*	1	WW	+	$1,2, \leftrightarrow$		-	\leftrightarrow	lz	l+	S-	1	1-	W	-	1	+	0	P-	f	fp↑	P1	-	PH±	4	a-	W	- 1	2	Pr+
Neğiş	1	g	±	_I<	S-	1	WW	+	1		-	0	lz	1-	S-	1	1-	W	-	1	-	0	P+	f	fp↑	P1	-	PH-	4	a±	W	-	1	Pr±
Plovdiv	1	g	-	1<	S-	1,2	WW,*	' +	1*		-	0	lz	1-	S-	1,2	I-	k	±	1	+	0,I	P+	f	fp↑	P1	-	PH-	4	a+	W	+	2	Pr+
Ranbelli	-	-	-	-	-	1	W	+	1	Π	Π	\leftrightarrow	sl	1-	s+	1,2	Π	W	±	1	+	0	P-	f	fp↑	P1	-	PH±	5	a±*	W	+	1	Pr+
Rocca	1	g	±	I<	S-	1	W	+	1	\cap	-	0	1z	l±	S-	1	1-	W	-	1	+	0	P-	f	fp↑	P1	-	PH+	4	a±	Μ	-	1	Pr+
Şumnu	-	-	-	-	-	1	W	+	0	-	-	0	1z	1-	S-	1	1-	W	-	1	+	0	P-	f	fp↑	P1	-	PH-	4	A±	W	-	1	Pr-
Trakya	1	g	±	<	s+*	1	WW	+	1*		-	0	lz	1+	S-	1	Π	W	±	1	+	0	P+	f	fp↑	P1	-	PH-	4	a±	W	-	1	Pr+
Tunca	1	g	+	Π	S-	2	WW	+	\leftrightarrow	\cap	\cap	$1, \leftrightarrow$	lz	1-	S-	1-2	l±	W	+	1	+	0	P+	f	fp↑	P1	-	PH+	4	a±*	W	-	1	Pr+
Yavuz	-	-	-	-	-	1	w	+	1	II	-	-	lz	1-	S-	1	1-	w	±	1	+	0	P-	f	fp↑	P1	-	PH±	5	a±	W	-	1	Pr+
Kızıltan	1	G	+	_I<	S-	1	WW	+	1*		-	-	lz	1+	S-	1	1+	W	+	1	+	0	P+	f	fp↑	P1	-	PH+	5	a+	WW	-	2	Pr+

Table 2. Anatomical properties of all cultivars in control (non-saline, Meriç) conditions. *: groups indicated in different colors as orange (Best), yellow (Good), green (middle) and blue (Low). For icon descriptions please refer to the Table 1 in Material and Methods section. For Endodermis "U" thickenings types, see Fig. 6a-g.

Table 3: Anatomical properties and plasticity of all cultivars grown in Salty Ergene field conditions. *: groups indicated in different colors as orange (Best), yellow (Good), green (middle) and blue (Low). Green, yellow and red areas indicate increasing, sameess and decreasing of the character, respectively. For icon descriptions please refer to the Table 1 in Material and Methods section. For Endodermis "U" thickenings types, see Fig. 6a-g.

Group*		Outer Exode			is	М	Middle Exod. Inner Exod.					Outer cortex, Aeranchyma Inner cortex Endodermis Per							Pericycle	/ Phloen	n		xylem													
	EOL	G	S	AW	' S+/-	EM	IL V	N	S	EIL	L	W	OC	AE	L	Si	IC	L	W	R	E	R	U ³	PS	P#	PH	PP	si	PH	Α	AS	М	Si	S	Pr	Prl
Kral	1	G	±	- I<	s+*	1	V	W	+	1	Π	\leftrightarrow	\leftrightarrow	lz	l±	S-	1	Π	W	+	1	+	Ι	P+	f-	fp↑	PO	-	PH+		a+	W±	-	-	1	Pr+
Kırkpınar	2	GG	+	- I<	S+**	1,2	2 V	N	+	1*	\cap	-	0	lz	1-	S-	2		W	+	1	+	Ι	P+	f	fp↑	PO	-	PH+		a+	WWW	-		1	Pr+
7721	1	G	+,	- I<	s+	1	, v	w	+	1*, s	Π	-	-	lz	1-	S-	2	-	W	+	1	+	Ι	-	f	fp↑	P0	-	PH-		a+	W	-	-	1	Pr+
Sürek	2	G	++	I<	s+**	1,2	2 x	w	+	0,↔		-	0,ss,+	lz	1-	S-	1		w	+	1	+	III	P+	p, L+	fp↑	P1	1, id	PH+		a±	W	-	L±, id	1	Pr+
Ece	1	G	±	I<	s+	1	X	w	+	1		-	1	1	l+	S-	2		∩,W	+	1	+	Ι	P+	ff	fp↑	P0	-	PH+, L+	▼	a+	W	-	-	1	Pr-
Kors-424	1	g	+	II	s+	1	7	w	+	1,*		-	0,↔	lz	1-	S-	2	\cap	-	+	1	+	III	P+	f	fp↑	P0	-	PH++	=	a+	W	-	-	2	Pr+
Gala	2	G	+	Π	s+**	- 1	V	N	+	1		-	\leftrightarrow	sl	1-	S-	1	L+	Π	+	1	+	Ι	P+	f, L+	fp↑	PO	-	PH+	=	a+	WW	-	-	1	Pr+, L+
Veneria	1	G	±	I<	S+	1	V	N	+	1, *	\cap	-	0	lz	1-	S-	1	L-	W	+	1	+	II	P+	f-	fp↑	P0	-	PH++	▼	a+	W	-		1	Pr+
Altınyazı	1	g	-	Π	S-	1	- V	W	+	1	Π	\leftrightarrow	\leftrightarrow	sl	l±	S-	2	L+	\cap	+	1	+	II	P+	f	fp↑	P0	-	PH+		a-	WW	-	-	1	Pr±,A
Durağan	1	G	+	<	s+**	- 1	- X	w	+	0	-	SS	1,B,+	lz	1-	S-	1,	L+	W	+	1	+	Ι	P++	f-	f=	P0	-	PH+		a+	W	-	-	1	Pr+
Halilbey	1	G	-	<	S-	1,2	2 V	W	+	1	Π	\leftrightarrow	\leftrightarrow	lz	1-	S-	1,2	1-	w	+	1	+	IV	P+	f	fp↑	P0		PH+	▼	a+	WW	-		2	Pr+,A
Koral	1	G	-	<	s+	1	- V	N	+	1		-		sl	L+	S-	1		w	+	1	+	IV	P+	f, L+	fp↑	P0	-	PH+	=	a+	WW	-	-	1	Pr+
N-41-T	2	G-G	+	_I<	s+	- 1	X	w	+	1, ↔	\cap	-	\leftrightarrow	lz	l±	S-	1,2		\cap	+	1	+	Ι	P+	f-	fp↑	P0	-	PH+		a±	W±	-		1	Pr+
Osmancık	1	G	+1	Π	s+*	1	`	w	+	1*	Π	-	0	lz	1-	S-	1	Π	k	±	1	+	IV	P++	F, L+	fp↑	PO	-	PH+	•	a+, W,+	W	-	S▲	2	Pr+,W,
Akçeltik	1	g	+	I<,	S-	1	`	w	+	0	Ω	\leftrightarrow	1	lz	1-	S-	1	L+	w	+	1	+	Π	p±	f	fp↑	PO	-	PH-	5	a	WW	-	Li▲, W▲	1	Pr+,A
Beşer	2	g	+	I<	S+	1,2	2 V	N	+	1	Π	-	-	lz	1-	S-	1	1-	W	±	1	+	Ι	P+	f	fp↑	P0		PH+		a-	WW	-	-	2	Pr±
Edirne	2	G-G	+	I<	S+	1	V	W	+	0	-	-	0	lz	1-	S-	1	Π	w	++	1	+	Π	P+	f	fp↑	PO	-	PH+	•	a+	WW	-	-	1	Pr+, L+
Gönen	1	G	±	- I<	s+	1	W	W	+	1			↔, B L+	, lz	1-	S-	1,2	Ω	w	+	1	+	IV	P+	F, L+	fp↑	PO	-	PH+	=	a-	ww	-	-	1	Pr+
İpsala	1	G	±	- I<	s+	1	X	w	+	1	$\cap \blacktriangle$	\leftrightarrow	\leftrightarrow	lz	1-	s+	1,2	L+	\cap	+	1	+	III	P+	f, L+, W	-	P1	-	-	▼	a-	WW -	-	Li▲	2	Pr+
Karadeniz	0	-	-	-	S+	1	- X	W	+,si	1	Π	\leftrightarrow	\leftrightarrow	lz	l-	S-	2	l+	W	+	1	+	Ι	P+	f-	fp↑	P0	-	PH-	▼	a±	W	+		0	Pr+
Kargı	2	G	-	<	s+	1	V	W	+	1**	Π	Π	0	lz	1+	S-	2		w	+	1	+	IV	P+	f, WW, L+	fp↑	P0	-	PH+		a-	ww	-	İd	1	Pr+,A, L+, W+
Meriç	2	G	+	<	s+*	1	τ.	w	+	0	-	-	0	lz	1-	S-	1	L+	w	+	1	+	Π	P±	f-	fp↑	PO	-	PH+		a	M-	-	-	1	Pr+
Neğiş	1	G	+	I<	s+	1	, s	w	+	-	-	-	\leftrightarrow	lz	l-	S-	1	\cap	w	+	1	+	III	P+	f-	fp↑	PO	-	PH±	=	a-	w	-		1	Pr±
Plovdiv	1	G	+	I<	s+	1	W	v*	+	1**			1	sl	l+	s+	1,2	Π	w	+	1	+	Ш	P+	f-, L+	fp↑	PO	-	PH+		a+	WW	-		1	Pr+, L+
Ranbelli	1	G	+	I<	s+	1	1	w	+	-	-	-	0	lz	1-	S-	1		\cap	+	1	+	III	P+	f	fp↑	P0	-	PH+	•	a±, L±	W-	+		0	-
Rocca	2	G	+	II	s+	1	1	w	+	-	-	-	\leftrightarrow	lz	l±	S-	1,2	\cap	w	+	1	+	III	P-	f	fp↑	P0	-	PH+, L+	=	a+	WW	-	Li▲	1	Pr±
Şumnu	1	G	-	- I<	s+	1	V	V-	+	0,↔	\cap	-	\leftrightarrow	lz	l-	S-	1		W	+	1	±	IV	P-	f	fp↑	P0	-	PH-	=	a±	WW	-	-	2	Pr-,W
Trakya	1	G	+	Π	s+**	- 1	W	W	+	0	-	-	\leftrightarrow	lz	l±	S-	2	Π	w	±	1	+	Ι	P+	f	fp↑	PO	-	PH±	=	a±,w,L ±	W	+		1	Pr±
Tunca	1	G	+	Π	s+	1	V	N	+	¢	Ω	-	0	lz	1-	S-	1	L-	w	+	1	+	III	P+	F, Li+, W	fp↑	PO	-	PH+, W	=	a- ,L±	WW	-	id	1	Pr+,W
Yavuz	1	G	+	I<	s+	1	V	N	+	1			\leftrightarrow	lz	1+	S-	2		W	+	1	+	IV	P+	F, W, L+	fp↑	PO	-	PH+, L+	▼	a±, L++	WW	-	L▲, id	1	Pr+, L+
Kızıltan	1	G	+	П	s+	1	V	N	+	1*	-	-	-	lz	1-	S-	2	1-	w	+	1	+	III	P+	f	fp↑	PO	-	PH++	•	a+	W	-	-	1	Pr+,L+, W

Table 4. Plasticity Strategies detected in different root tissues in the salty area. #: shows the alternative strategies, +: shows additional character as well as said character, /: shows different forms of the same character.

Best	E: Increasing layering + silica, wall thickness, lignification, (altogether). C- ED: IC: layering + IC wall thickness + lignification degree; aeranchyma lignified or not; ED, "Uth" feature. P- PH: P interruption (degree), PH: cell layout, PH gap scarcity / abundance. Xy: A , number increase / unchanged + solid, robust / partially damaged. M , area increased. Prt , kept its density and normal thick-walled lignified feature.
Good	E: wall thickness, Lignification density, cell size, alternating with cortex or not, layering. C-ED; OC: unchanged # appearing in saline cond.; Aeranchyma: lizigen to Schizo-lizigen # lignification decrease / increase; IC: layering + regularity + wall thickness, altogether, increased # layering unchanged but regularity + lignification increased # layering unchanged + regularity increased but non-lignified. ED: "Uth" More regular and thickneed # partially thickened + thin-walled # slightly thickened, damaged # thinned, uncertain # unchanged, thin-walled. P-PH: P: interruption degrees / P loss; cells more regular # cells lignified # not interrupted continuous + idiyoblast and silica deposition present + lignified fairly. PH: with gaps # more regular / irregular # partially lignified. Xy: A , number increased/unchanged. Tracheal and xylem's solid structure present. M, layout unchanged / partially reduced + silica lost # Silica generally present. Prt, unchanged in density # unchanged in density but lignification lost / increased # kept fairly regular thin-walled, lignified structure # kept its damaged state # kept i
Middle	E : cell size, strength, alternating with cortex or not, degree of lignification, wall thickness. C - ED ; OC : more regular, large # partially evident # absent by alternating with E. Aeranchyma partly schizohenic, lignified (back to full lignified) # partially schizogenic, full lignified # both lizigen, non-lignified # both lizigen and partially lignified. IC : layering unchanged / increased + outer periclinal wall (OPC) thicker + entire wall lignified # layering unchanged + all wall surface lignified # layering unchanged + OPC lignified # layering unchanged + non-lignified wall. ED : "Uth" quite evident / thickened # partly evident / thickened (little) # uncertain # always uncertain, unchanged. P - PH: P: discontinuity unchanged / decreased # discontinuity present + some cells lignified and more regular # gaps absent. PH: unchanged/more regular. Xy: A , number increased + walls partially damaged + silica lost # increased + Xy more regular # number unchanged / decrease # wall's damages absent + Xy more regular + lignification increased. M , quite thick-walled # wall thickness partly increased # wall thickness reduced + walls damaged. Prt , unchanged in normal density, lignified and robust feature # integrated with A + low density + thin-walled + less lignified # integrated with A + walls thicker and lignified # declined in frequency + maintained its regular lignified thickenings # kept its intensity + kept thick-walled and same lignified state.
Low	E; EO layering increased / unchanged/increased + dense/low silica # layering decreased + lignified thick-wall # cells larger/completely lost + silica increased. EM layering + lignification + wall thickness decreased # layering unchanged + lignified wall thinned / thicknend. EI layering decreased + unchanged thick-walled lignification # layering + walls unchanged # alternated with OC # entirely lost. Layering, silica, lignification, wall thickness, alternation or losing in several combinations found. C-ED; OC: layering decreased # lost # partly present and widened / lignified # formerly absent, subsequently alternating with Ex # more impaired. Aeranchyma partially returned from schizolizigen to lizigen # non-lignified # increased lignification # unchanged, non-lignified # silica increased/never. IC: layering increased, but lignification unchanged # layering increased + lignification = wall thickened # layering unchanged + lignification spread over the entire wall surface / partially increased / lignification lost # layering unchanged + lignification increased/decreased/partially emerged / lignification lost # layering walls thickness. E, returned as follows: little more evident, but not too thick # thicker and more regular # quite evident-thickned # unchanged form # lignification and wall thickness increased. PH: P: discontinuity unchanged/decreased + silica lost + wall damage dense / regular-robust # unchanged + trache heavily damaged + silica lost # unchanged + silica increased + wall damage dense / regular-robust # unchanged + trache neavily damaged + silica indense + lignification increased + wall damage dense # decreased + lignification increased # decreased # not lignified # damages dense # decreased + lignification increased # decreased # wall damage dense / regular-robust # unchanged + trache heavily damaged = silica lost # unchanged + silica increased # wall damage partly few/increased # decreased in density + integrated with A # increased + lignification increased # decreased # not lignif

		Parametre number*												
Group	Exodermis	Cortex Endodermis	Pericycle Phloem	Xylem	Total									
Best	4	5	3	5	17									
Good	5	15	8	15	43									
Middle	5	20	7	15	47									
Low / very low	7	30	8	25	70									

Table 6. Si contents (mg/kg) in cultivars grown in control and salty conditions. *: Meriç and Ergene respected control and salty conditions, respectively.

Kıral 540.39 543.7 Kırkpınar 366.78 479.46 Sürek-95 249.54 610.87 7721 495.37 425.5 Ece 436.56 678.34 Kros-424 463.03 319.34 Gala 429.07 562.14 Veneria 398.32 472.97 Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz <t< th=""><th>C. H</th><th></th><th>Si*</th></t<>	C. H		Si*
Kırkpınar 366.78 479.46 Sürek-95 249.54 610.87 7721 495.37 425.5 Ece 436.56 678.34 Kros-424 463.03 319.34 Gala 429.07 562.14 Veneria 398.32 472.97 Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 <th>Cultivars</th> <th>Si* (Meriç)</th> <th>(Ergene)</th>	Cultivars	Si* (Meriç)	(Ergene)
Sürek-95 249.54 610.87 7721 495.37 425.5 Ece 436.56 678.34 Kros-424 463.03 319.34 Gala 429.07 562.14 Veneria 398.32 472.97 Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 59	Kıral	540.39	543.7
7721 495.37 425.5 Ece 436.56 678.34 Kros-424 463.03 319.34 Gala 429.07 562.14 Veneria 398.32 472.97 Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halibey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65	Kırkpınar	366.78	479.46
Ece436.56678.34Kros-424463.03319.34Gala429.07562.14Veneria398.32472.97Osmancık477.66826.3N-41-T299.6584.66Durağan327.48666.11Koral357.5371.95Altınyazı276.7516.87Halibey398.68523.09Neğiş683.09373.45Trakya616.32452.41Şumnu191.5548.86Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Sürek-95	249.54	610.87
Kros-424463.03319.34Gala429.07562.14Veneria398.32472.97Osmancık477.66826.3N-41-T299.6584.66Durağan327.48666.11Koral357.5371.95Altınyazı276.7516.87Halibey398.68523.09Neğiş683.09373.45Trakya616.32452.41Şumnu191.5548.86Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	7721	495.37	425.5
Gala 429.07 562.14 Veneria 398.32 472.97 Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361	Ece	436.56	678.34
Veneria 398.32 472.97 Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 42	Kros-424	463.03	319.34
Osmancık 477.66 826.3 N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Gala	429.07	
N-41-T 299.6 584.66 Durağan 327.48 666.11 Koral 357.5 371.95 Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Veneria	398.32	472.97
Durağan327.48666.11Koral357.5371.95Altınyazı276.7516.87Halilbey398.68523.09Neğiş683.09373.45Trakya616.32452.41Şumnu191.5548.86Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Osmancık	477.66	826.3
Koral357.5371.95Altınyazı276.7516.87Halilbey398.68523.09Neğiş683.09373.45Trakya616.32452.41Şumnu191.5548.86Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	N-41-T	299.6	584.66
Altınyazı 276.7 516.87 Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Durağan	327.48	666.11
Halilbey 398.68 523.09 Neğiş 683.09 373.45 Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Koral	357.5	371.95
Neğiş683.09373.45Trakya616.32452.41Şumnu191.5548.86Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Altınyazı	276.7	516.87
Trakya 616.32 452.41 Şumnu 191.5 548.86 Gönen 240.47 558.02 Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Halilbey	398.68	523.09
Şumu191.5548.86Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Neğiş	683.09	373.45
Gönen240.47558.02Kızıltan388.11542.21Beşer708.37812.8Kargı439.11368.09Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Trakya	616.32	452.41
Kızıltan 388.11 542.21 Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Şumnu	191.5	548.86
Beşer 708.37 812.8 Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Gönen	240.47	558.02
Kargı 439.11 368.09 Edirne 384.02 318.94 Karadeniz 208.83 640.28 Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Kızıltan	388.11	542.21
Edirne384.02318.94Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Beşer	708.37	812.8
Karadeniz208.83640.28Rocca286.2429.43Yavuz596.6479.11Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Kargı	439.11	368.09
Rocca 286.2 429.43 Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Edirne	384.02	318.94
Yavuz 596.6 479.11 Ranbelli 45.69 - Meriç 331.26 528.65 Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Karadeniz	208.83	640.28
Ranbelli45.69-Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Rocca	286.2	429.43
Meriç331.26528.65Plovdiv762.96551.92Akçeltik361.21526.04Tunca427.4470.87	Yavuz	596.6	479.11
Plovdiv 762.96 551.92 Akçeltik 361.21 526.04 Tunca 427.4 470.87	Ranbelli	45.69	-
Akçeltik 361.21 526.04 Tunca 427.4 470.87	Meriç	331.26	528.65
Tunca 427.4 470.87	Plovdiv		551.92
• • • • • • • • • • • • • • • • • • • •	Akçeltik	361.21	526.04
İpsala 407.42 -	Tunca	427.4	470.87
- F	İpsala	407.42	-

Discussions

General anatomical structure: The epidermis was always absent, with 1–2-layered outer exodermis, 1–2layered sclerenchymatous middle exodermis, 1–2-layered inner exodermis, outer cortex present or absent, sometimes alternating with inner exodermis, lysigenous or partially schizo-lysigenous aerenchyma and inner cortex subtending to 1 row of endodermis and pericycle in stele, 3–6 arcs of xylem consecutive with the phloem. The phloem was sometimes cut intermittently by the pericycle, with the xylem parenchyma present. The medullary structure was thick-walled and lignified. Apoplastic barriers were scattered in different regions of the root, depending on the cultivar, such as sclerenchymatous middle exodermis and endodermis. Sometimes the pericycle, outer and / or inner cortex as well as the aerenchyma became lignified. Similarly, wild rice Zizania latifolia, a close relative of rice, showed suberised and even lignified endodermis (stage III) in response to salt. Adjacent, thick-walled cortical layers, single-layered suberised-lignified sclerenchyma layer, lysigenous aerenchyma and thick-walled inner cortical cells were also found in wild rice. In general, apoplastic barriers are present variously in roots (Fleck et al. 2011, Chaodong et al. 2014) For example, Z. aquatica, which is also closely related to rice, shows an uniseriat exodermis and biseriate hypodermis (Clark & Harris 1981, Kotula et al. 2009) and the epidermis is often excised. This lack of root epidermal tissue is not uncommon in Poales (Rebouillat et al. 2009, Chaodong et al. 2014). Regarding the aerenchyma, a schizo-lysigenous to lysigenous state in the stem and leaf cortex of wild rice is normal and similar Cynodon dactylon, Eremochloa ophiuroides, to Hemarthria altissima and Miscanthus sacchariflorus (Yang et al. 2011). These air spaces transport oxygen to organs under hypoxic conditions (Chaodong et al. 2014), a crucial role of aerenchyma in aquatic and amphibious plants (Chaodong et al. 2011).

For all the groups (Best to Low), the response of each of the anatomical region to salinity stress are separately discussed below.

Exodermis: In the Best group, essential or strategic parameters only appeared in the Kırkpınar cultivar and Kral cultivar showed almost no difference. Here other possible physiological parameters (ions, hormones, antioxidants, osmolytes, etc.) were activated. Indeed, the yield of Kırkpınar was reported to be higher than that of Kral (Aybeke & Demiral 2012). The essential anatomical parameters given in Table 5, illustrate the important protective traits for these plants. In the Good group, cell size (large and robust) was an additional parameter essential for salt tolerance. Under normal circumstances (i.e. non-saline conditions), these cells were tiny and crushed or damaged but under saline stress they were large and more uniform. The yield values of these largeand regular-celled cultivars (e.g. 7721, Gala, Veneria) were reported to be higher than those of other cultivars in the Good group (Aybeke & Demiral 2012). In the Best group, this parameter was not observed as a variable, because in this group, the cells were always regular. In addition, Si was also increased and dense suberised material was observed (Fig. 7c). I believe that Si and suberin accumulation in the outer exodermis increased

resistance to salt stress. In addition, in the Good group, these essential parameters showed inverse trends of increase or decrease. However, all these parameters increased or decreased together in the Best group. In the Middle group, the exodermal layer number was important, along with the state of alternation with the cortex. However, this status was not normally seen in the Best group and in some of the cultivars in the Good group. Accordingly, the exodermal row number was replaced by alternation state as a protective anatomical structure. In the Low group, row number (decrease / increase / absence / equal), thick and lignified wall status and presence / absence of Si emerged as more variable parameters. In summary, the following results were observed: (1) All essential parameters were at the highest levels in the Best group, (2) the best protection was achieved by changes directed towards higher level of protection or keeping tissue components at the higher level, (3) any decrease in these parameters was accompanied by the addition of new parameters to resolve protective gaps, (4) in the Good, Middle and Low groups, the 'essential' parameters were lacking, decreasing or varying inversely with each other and (5) row number was an important parameter and lignified thick wall and cell-size regularity increased strength with Si. Accordingly, it was concluded that the exodermis is an important apoplastic barrier for resistance to salt stress.

Cortex and endodermis: In the Best group, the overall cellular changes in the inner cortex and endodermis of Kırkpınar were seen to increase substantially but such changes in the Kral cultivar remained to be limited (Table 3). Kırkpınar cultivar showed more effective protection with new anatomical changes in its cortex, endodermis and exodermis. In other groups (Good, Middle and Low), different parameters such as the robustness/disorder of the outer cortex, alternating status, aerenchymatous features, lignification degree, presence/absence of inner cortex, row number, lignification, wall features and endodermal thickening changed inversely with each other. There were more parameters in these groups than in the Best group because resistance to salt has shifted inwards to the inner cortex and even to endodermis. Therefore, if parameters (such as the exodermis) are not effective, additional traits would provide salt resistance. Similarly, the inner exodermis and outer cortex in Ranbelli, Rocca, Şumnu, Trakya and Tunca cultivars were very severely damaged. In these cultivars, increasing protective changes occured in the outer exodermis, inner cortex and to some extent in the endodermis. In a recent study on drought and roots, large xylem vessels with lesser aerenchyma formation and higher starch content in tolerant rice varieties were required for the maintenance of water potential and energy storage (Singh et al. 2013). However, in the present study, starch was not found. Under saline conditions, the starch content in rice roots declined (Paridaa et al. 2005). I believe that absence of starch in fully developed aerenchyma lacking a cellular layout is a natural consequence of this response to salinity.

Pericycle and phloem: In all the groups, interruption ratio of the pericycle and its cell layouts, lignification status, features of the phloem and distributions of phloem gaps emerged as the main parameters. In short, under saline conditions, the pericycle's layout was improved, lignification increased and parenchyma and phloem became more regular. Even idioblastic structures, Si deposition and lignin formation were seen in the pericycle and phloem. Based on these parameters, although all cultivars exhibited tremendous variation, a fundamental and significant variable that can determine groups in terms of 'pericycle and phloem' could not be identified. Therefore, anatomical properties of the pericycle and phloem cannot be used for distinguishing salt-resistant specimens.

Xylem: The arc number increased or just remained the same, additionally the arc integrity, medulla widening, protoxylem density, lignification and degree of wall thickness were important and constant parameters. Furthermore, regularity of the xylem parenchyma was also an important variable. Indeed, recent data indicate that within the stele, pericycle and xylem, parenchyma cells are important in the control of the net Na⁺ flux of the xylem (Koyro 2002, Läuchli et al. 2008). Even though no relationship between arc number and salt resistance was found, I believe that an increasing arc number will dilute the passage of apoplastic salt-water mixture because of the increase in the total water intake of the plant. Thus, the plant will reduce toxicity of the salt it takes in. Similarly, Sobrado (2007) found that generally, higher salinity may result in narrow vessels and increase vessel density, maximising water uptake under high-salt conditions. Therefore, it was logically understood that organization of the vascular structure is an individual important parameter in breeding, as indicated by Shigenori & Nemoto (1995).

When changes were analyzed according to the root region and group (Table 5), the minimum value belonged to the Best group, particularly in the Kırkpınar cultivar, for which all essential parameters were used. On the contrary, in other groups, because of a decline in essential parameters, other new variables were activated and the number of parameters increased gradually till the Low group. The number of parameters was quite high in the exodermis, cortex-endodermis and xylem but in contrast, the number of parameters in the pericycle and phloem was the lowest. Because the phloem, pericycle and parenchyma are meristematic, with living, dividing and specific tissues, wall thickening by lignification cannot be implemented as a valid strategy. Indeed, the essential parameters remained stable from the initial phase into a continuing period of stress. The response of roots to stress do not require both other 'essential variables' and 'additional parameters'; instead the initial variation is sufficient and only the essential parameters are required. I believe that this anatomical stability should increase plant efficiency by saving energy or else the higher variability will cause lower plant yield, as demonstrated by the current study. Based on these findings, the xylem, cortex, endodermis and

exodermis were considered 'essential strategic' areas (apoplastic barrier) providing the highest anatomical resistance. In addition, the degree of root anatomical plasticity and stability (more or less) under stress conditions is undoubtedly another important concept as described above.

In the Low group cultivars such as Kargı, Yavuz, Plovdiv, Rocca, İpsala and Akçeltik, dense and thickwalled lignifications, phenolic accumulation and idioblast cells filled with gummic structure were found to be densely arranged in stele rather than exhibiting an improvement of essential parameters in roots. Similar results were recorded as vascular / intercellular blockages induced in roots or rhizomes of plants by wounding, containing polysaccharide gums, callose, beta-1,3-glucan polymers (Soukup et al. 2002). As a result, vascular blockages induced by phytotoxins or environmental stress and other lignified blockages eventually developed in the metaxylem in all the protoxylem and even in the phloem. Votrubová et al. (1997) and Soukup (1997) reported that these blockages contain polysaccharide gums derived from non-cellulose wall components. Thus, both water and useful / harmful ions transported via the apoplastic pathway may be impeded by these growths. Consequently, the underground system suffered from a shortage of all useful metabolites, affecting growth, food storage and the plant's capacity (Sánchez-Aguayo et al. 2004, Armstrong & Armstrong 2005). Therefore, the present findings support previous work demonstrating that lipids, suberin and sometimes only lignin-type phenolics may increase lignin/phenolic contents resulting from alterations in plant secondary metabolism following oxidative stress (Schutzendubel & Polle 2002), and such additional abnormal changes in vascular structure can decrease yield (Aybeke & Demiral 2012). Bualuang et al. (2012) also found that under prolonged salt-stress conditions, as in present study, sufficient development of the apoplastic barriers of the outer protective tissue lowers the magnitude of bypass flow, increasing seedling survival. However, the authors did not indicate how much apoplastic barrier development is sufficient to induce this effect nor they characterize the apoplastic barriers. In the present study, these gaps were filled entirely.

The endodermis is an important part of the root apoplastic barrier, with lignified–suberised Casparian strips that block apoplasmic continuity in the pathway between the cortex and stele. Solutes then crossed the endodermis via passage cells within this layer and thus traverse a plasma membrane. Plant membranes, in general, have low permeability to both Na⁺ and Cl⁻ ions so that the endodermis, with its Casparian strips, probably restricts the flow of Na⁺ and Cl⁻ ions to the stele (Atwell et al. 1999). As in present study, the endodermis showed significant structural 'U' thickening improvement under saline conditions and even reduced the density of Si ions (which is useful for enduring salt stress) within the vascular system. Indeed, the endodermis is an important protective apoplastic barrier against ionic stress, as shown in Table 2–4; Fig. 6a-g, 7b, 7d.

Silicon is a useful element in soil that effectively counteracts the effects of various abiotic stresses such as drought, heavy metal toxicity and salinity (Hashemi et al. 2010). Partial substitution of lignin by silicon or formation of silicon-polyphenol complexes in walls may facilitate wall loosening and promote the growth of plants under stress conditions. In addition, silicon accumulation showed an increasing in cell wall extensibility in sorghum roots (Hattori et al. 2003) and caused an increasing in N, P and K uptake. Kaya et al. (2006) observed that silicon application increased the contents of Ca and K in maize under water stress, whereas Chen et al. (2011) found that silicon supplementation decreased the contents of K, Na, Ca, Mg and Fe in rice as well as reduced chloride transport in rice plants by reducing transpirational bypass flow. Silicon generally increased in all cultivars under saltstress conditions and showed no significant and specific difference among the groups (Table 6). Si contents were similar in the exodermis and endodermis in Low group cultivars (i.e. Akçeltik), whereas in the Best group (Kırkpınar), the content decreased gradually towards the endodermis and vascular system. Moreover, the increase in Si content and wall thickness in Kırkpınar positively influenced yield values. In our previous study (Aybeke & Demiral 2012) Na content was high and based on present results, increasing Si content did not reduce the Na content, the, contrary to the findings of Chen et al. (2011). Thus, these conflicting results raise doubts regarding the effectiveness of Akceltik endodermis apoplastic barriers and passage cells electivity. Although the endodermis of Akçeltik (Type II, Fig.6e) is thicker than Kırkpınar's (Type I, Fig. 6d), understanding this pattern will require an additional physiological study on endodermis cells. Thus, based on this interpretation, it is clear that not only endodermal thickness but also endodermal activity is an important element in response to salt stress. In addition, no study has compared Si distribution in the roots of saltresistant and susceptible rice cultivars. Therefore, these findings are new and important to prepare the ground for new work and reveal the relationship between Si content and apoplastic barrier.

Recent studies (Henry et al. 2012) did not find structural modifications associated with adaptation to saline / drought environments, inferring that physiological regulation may be more important. However, according to recent detailed studies (Enstone et al. 2003, Henry et al. 2012), rice cultivars differing in salt-stress resistance may be used to select or create new varieties of crops to obtain better productivity under salt-stress conditions and to thoroughly understand the morpho-anatomical and physiological basis of salt-stress resistance. Because rice has a complex resistance mechanism (morphological, biochemical and physiological) in response to salt stress and anatomical parameters are also to beat least as preferable as those of other ones (Enstone et al. 2003, Henry et al. 2012). Therefore, this information will undoubtedly enable genetic improvement of salt resistance in rice and will provide potential benefits to breeding studies (Mostajeran & Rahimi-Eichi 2008, Suralta & Yamauchi 2008). In addition, genotypic variations under salt-stress conditions and root anatomical adaptation selection parameters will be important in a successful selection and breeding of new cultivars (Hashemi et al. 2010, Krishnamurthy et al. 2011, Henry et al. 2012). Furthermore, root anatomy is directly related to tolerating other environmental stresses. Thus, several root anatomical traits have potential for use in crop breeding. Similar findings were emphasized in a previous study (under full-season and real field conditions) related to Na, K, Ca ionic parameter changes (Aybeke 2016). Resistant cultivars have exhibited rapid and adequate responses as part of physiological adaptations from the normal (nonsaline) conditions to stress conditions (Aybeke & Demiral 2012, Suralta et al. 2008).

Lynch et al. (2014) stated that full-season real field experiments will be important tools in concert with empirical studies and this sort of challenge will require expertise in plant biology, requiring renewed emphasis on understanding the plant phenome. Because of the complexity of the soil environment and the large number of potential interactions and possible scenarios, functional-structural plant studies that focus on topics such as root anatomy will become important. A more challenging obstacle is the need to understand the utility of specific root phenotypes in the context of specific agroecosystems and specific phenotypic and anatomic backgrounds, namely the 'fitness landscape' (Lynch & Brown 2012). Accordingly, this type of study, a 'fullseason and real field' experiment will facilitate the utility of any given root phenotype for drought, salinity or other stress tolerance. Chen & Wanga (2009) stated that creation of anatomy-based screening techniques is possible only after long-term natural stress conditions because plant adaptation mechanisms only mature in long-term field conditions. In addition, Lynch (2014) stated that suberization and lignification should become selection targets in plant breeding. However, much more is needed to be learned about the environmental conditions and development of apoplastic barriers in various root tissues. For instance, what are the advantages

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and disadvantages of earlier and/or more intense suberization of root endodermal, exodermal and sclerenchyma layers under stressed and unstressed conditions and whether plasticity of these traits will be advantageous or not? The present study clearly provided answers to these questions. In addition, the simplicity, ease, economic feasibility and speed of obtaining anatomical results are other important advantages of the current method. In future studies, more detailed physiological mechanisms of the roots in the Best group, the relationship between aquaporin expression and salt resistance, antioxidant mechanisms and the interaction between apoplastic barrier development and Si will be examined in the context of other physiological and hormonal changes. Thus, the morphological, anatomical, physiological and genetic basis of salt resistance will be examined thoroughly and new, reliable and reproducible techniques will be created for identifying more resistant rice varieties.

Conclusion

In the present study performed in order to reveal detailed root anatomical parameters that can be used to select and breed salt-tolerant rice, root anatomical plasticity of several rice cultivars (from Best to Low vielding) were investigated under saline and non-saline field conditions. According to the overall results obtained, the salt resistant group (Best) showed that protective anatomical characters improved or remained equal to those of the control, but in other groups (Good, Middle, Low), these parameters were found as worsened or remained equal to those of the control. Although anatomical plasticity is essentially directly related to apoplastic barrier features, nevertheless cell size, elemental distribution of Si, xylem properties, lignification-suberization degrees on apoplastic barriers, presence/absence of idioblast cells are determined as important root anatomical data for salt-resistant specimen selection. Additionally, it was understood that cultivar(s) bearing the most stabile anatomy under saline and nonsaline conditions was favorite one to select and breed saltresistant rice. Moreover, the simplicity, ease, economic feasibility of the current method was also highlighted as other important advantages.

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