Differential responses of roots for varying tolerance to salinity stress in wheat with special reference to elasticity

Yang Shao[1], Ping An[1] *, Xiaohui Feng[2], Muhammad Irshad [3],Victoria Otie [4], Weiqiang Li[5], Yuanrun Zheng[6], Yunus Qiman[7]

[1]Arid Land Research Center, Tottori University, 1390 Hamasaka, Tottori city 680-0001, Japan

[2]Key Laboratory of Agricultural Water Resources, Center for Agricultural Resources Research, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, 286 Huaizhong Road, Shijiazhuang 050021, Hebei, China

[3]Department of Environmental Sciences, COMSATS Institute of Information Technology, University Road, Abbottabad, Pakistan

[4]Department of Soil Science, Faculty of Agriculture, Forestry and Wildlife Resources Management, University of Calabar, P.M.B. 1115, Calabar, Nigeria.

[5]Signaling Pathway Research Unit, RIKEN Center for Sustainable Resource Science, 17-22, Suehiro-cho, Tsurumi, Yokohama 230-0045, Japan

[6]Key Laboratory of Resource Plants, Institute of Botany, Chinese Academy of Sciences, 20 Nanxincun, Xiangshan, Beijing 100093, China

[7]College of Forestry and Horticulture, Xinjiang Agricultural University, 311 Nongdadong Road, Urumqi 830052, Xinjiang, China

*Corresponding author. E-mail: an.ping@tottori-u.ac.jp

Acknowledgements

This study is partially funded by the Grant-in-Aid for Scientific Research (C) of Japan Society for the Promotion of Science, No. 26450020.

Abstract

Two salt-sensitive (Yongliang-15, GS-6058) and two salt-tolerant (JS-7, Xinchun-31) wheat cultivars were used to investigate the extension, extensibility (viscoelastic parameters), and chemical composition of the cell walls in their root elongation regions (apical 10 mm-long root segments), under salinity stress. The elasticity of the root cell wall, indicated by E_0, significantly decreased in the salt-sensitive cultivars, whereas the E_0 in the salt-tolerant cultivars was maintained at the same level as that in the non-saline condition. Root extension and the differences among cultivars were largely dependent on elastic extension, which accounts for one-half to two-thirds of the total extension. Viscosity, indicated by η_0, and the plastic extension of the root cell walls did not change across the treatments and cultivars. The significant decrease in cell wall elasticity in the root elongation region was one of the factors that depressed root growth in salt-sensitive cultivars under the saline condition. The well-maintained elasticity of salt-tolerant cultivars alleviated the depression of root growth by NaCl. Cell wall elasticity was positively correlated with the relative pectin and hemicellulose I contents and negatively correlated with the relative cellulose content. Under saline conditions, the relative hemicellulose II content did not change in the salt-sensitive cultivars; however, it decreased in the salt-tolerant ones. Thus, changes in chemical composition of the cell wall were correspond with the cell wall extensibility and root growth in wheat cultivars with different degrees of salt tolerance.

Keywords: Root elongation, Cell wall loosening, Apical root, Creep, Cultivar difference, Salt stress

Introduction

Plant cell wall elasticity describes the elastic properties of the wall polymers. When sustained force is applied to a cell wall, such as turgor pressure, the stretch in the wall is partly elastic and partly plastic (Boudaoud, 2010; Monlia, 2013). These terms refer to time-dependent extension. These extensions are results of the polymeric nature of plant cell walls (Cosgrove, 2018).

Cell wall extension, composition, structure, and growth dynamics have been extensively reviewed by Cosgrove (2018). The primary cell wall behaves like a viscoelastic composite material that demonstrates a time-dependent extension under load and time-dependent shrinkage after stretching (Boudaoud 2010; Cosgrove 2018). Modules E_0 and η_0 are the most significant parameters that indicate the elastic and viscous properties of the root-cell-wall, respectively.

Changes in the cell wall composition in relation to the cell wall extensibility have been reported. Pectin and de-esterification of pectic homogalacturonan have been associated with wall stiffening and growth cessation (Siedlecka et al. 2008; Hongo et al. 2012; Wang et al. 2020). Decreases in the amount and molecular mass of hemicellulose have been shown to increase the cell wall extensibility in the azuki bean (Kaku et al. 2002), tea roots (Safari et al. 2018), tomato hypocotyls (Miedes et al. 2011), and Arabidopsis (Xiao et al. 2016). A denser assembly of cellulose microfibrils induces wall stiffness (Podgórska et al. 2017). A rice mutant with a defect in root elongation showed a significantly low extensibility and high cellulose and hemicellulose II contents in the root-cell-wall in the elongation zone (Inukai et al. 2012). Collectively, these previous reports indicated that the chemical composition and extensibility of the cell wall inherently interact and sensitively respond to the growth environment.

The effects of abiotic stresses on the root-cell-wall extension are fairly limited.

Water deficit has been shown to reduce the cell wall extensibility of the root elongation zone in maize (Fan et al. 2006). Excessive aluminium (Al) in culture media depressed cell wall extension in the root apical zone in wheat (Tabuchi and Matsumoto 2001; Ma et al. 2004). Application of silicon (Si) increased the elastic extension and viscocity of the apical root-cell-wall in sorghum under drought conditions (Hattori et al. 2003). Compared to roots, the hypocotyl and leaves have been more extensively studied. Water deficit reportedly decreased the cell wall extensibility in the hypocotyl of soybean (Wu et al. 2005), and drought stress decreased the cell wall elasticity in rose leaves (Al-Yasi et al. 2020). Si application was shown to increase the leaf cell wall extensibility in rice, oat, and wheat seedlings (Hossain et al. 2002), and lead exposure reduced the leaf cell wall extensibility in rice (Hossain et al. 2015). NH_4^+-toxicity reportedly increases the cell wall rigidity, which limits the expansion of leaf cells (Podgórska et al. 2017). Auxin has recently been found to stimulate cell elongation by increasing the wall extensibility (Barbez et al. 2017; Majda and Robert 2018). Abiotic stresses seem to generally depress the cell wall extensibility; however, the effects of salinity (Na^+ ions) on root-cell-wall extension and extensibility have not yet been reported.

Under saline conditions, higher proportions of pectin and lower proportions of cellulose have been associated with cultivar differences in root growth in soybean (An et al. 2014a). The widely reported elevation effect of calcium (Ca) application on root growth under salinity stress was partially attributable to enhanced pectin levels (An et al. 2014b). A lower proportion of wall cellulose in the hypocotyls of squash and cultured tobacco cells ameliorated the inhibition in cell expansion and elongation under salinity stress (Sakurai et al. 1987; Iraki et al. 1989). The structural arrangement of cellulose microfibrils was altered by salt exposure in sorghum (Koyro 1997). The amount of cellulose in the primary root was shown to decrease in response to salinity stress in cotton (Zhong and Lauchli 1993) and soybean (An et al. 2014a). In *Artemisia annua*, the main changes in the cell wall were found in the structure of pectin under salt stress (Corrêa-Ferreira et al. 2019). Feng et al. (2018) reported that salinity damaged the cell walls in Arabidopsis by disrupting pectin crosslinking. Wang et al. (2020) reported that sodium induced pectin de-esterification, which reduced cell wall stiffness in isolated onion epidermel cells. The extension coefficient of wheat leaves was decreased even under short-term salinity exposure (Veselov et al. 2009). While the genes encoding xyloglucan-related enzymes, which are functional in the enhancement of root growth, were upregulated under long-term salinity exposure (Mahajan et al. 2020).

However, cultivar differences in root growth in relation to the cell wall extensibility, extension, and compositions in crops have not been reported previously. Therefore, the present study investigated the root-cell-wall extension parameters and extension and chemical compositions in the elongation region of young wheat seedlings under saline and non-saline conditions.

Materials and methods

Cultivation of wheat seedlings

Based on the growth and yield of the cultivars grown in saline soils in the northwest of China (personal communication with local researchers), two salt-sensitive (Yongliang-15, GS-6058) and two salt-tolerant (JS-7, Xinchun-31) wheat cultivars were selected as the experimental materials. Seeds of the four cultivars were surface sterilised in 5% sodium hypochlorite (NaOCl) for 5 min and then rinsed with distilled water three times. Twenty seeds were placed in a line on a sheet of filter paper. Each prepared sheet of filter paper (with the wheat seeds) were placed in a 24 × 34 cm plastic zipper bag and moistened with distilled water. The plastic bags containing the seeds were vertically placed in growth chambers (SANYO MLR-350 HT, Japan) set at 25 °C. Two days later, when the roots and leaves had reached lengths of ~1.5 cm and 1 cm, respectively, 80 mM NaCl (which gives distinct cultivar differences in root growth) solutions with 1/12 fold of Hoagland solution were reinsed on roots everyday. The same solution without NaCl was used as the control (non-saline treatment). Excess solutions were drained. During the treatment period in the growth chambers, plants were exposed to light (2000 lx) conditions of 16/8 h (light/dark) and temperatures of 23/18 °C (day/night).

The lengths of all primary and seminal roots (usually three roots were generated from one seed) in four randmly selected bags of each treatment were measured daily. Since the primary and seminal roots had similar length so the everage length of all roots in one bag was taken as one replicate for root length measurement. Ten days after the NaCl treatments, when there were significant differences in root length between the sensitive and tolerant cultivars in the 80 mM NaCl treatment group, roots of the seedlings were sampled for extension and chemical composition analysis.

Measurement of root-cell-wall extension

The extension of the apical root cell wall was determined following the method developed by Tanimoto et al. (2000). For each treatment, 30–50 roots from 4–5 growth bags were measured. Sections of the root region between 3 to 8 mm behind the root tip (5 mm-long section) were subjected to the extension measurements using a Creep meter (Yamaden RE2-3305C, Japan). A tensile force of 0.05 N was found to be optimal for obtaining typical clean and stable creep extension curves for these wheat roots. Elastic parameters (E_0, E_1, E_2, E_3), the plastic parameter, the viscosity coefficient (η_0, η_1, η_2, η_3), and the total, elastic, and plastic extension distances were determined by the computer program, based on the Kelvin-Voigt-Burgers model (Tanimoto et al. 2000).

Measurement of chemical compositions in the root-cell-wall

Roots were taken out from growth bags and were rinsed with distilled water three times, and then 10 mm-long apical segments were excised with a razor blade. Root samples from 4–5 bags containing ~160 root segments represented one replicate and 4–5 replicates were taken per treatment. The fresh weights of these segments were recorded. Some segments were assigned for dry weight measurement, i.e. placed in an oven set at 90 °C for 3 days prior to measurement. The water content of all cultivars under the control and salinity treatments were calculated. Based on the water content, the dry weights of the segments were calculated to determine the composition measurements. Cell wall compositions were analysed using the procedure of Zhong and Lauchli (1993) with minor modification. Specifically, root segments were homogenised with ice-cold Tris-HCl buffer (pH 7.4) and Tris buffer-saturated phenol using a µT-12 bead crusher (Taitec Corporation, Koshigaya, Japan). The homogenate was centrifuged with 15 minutes, 5 000 g at 10 °C. The supernatant was discarded and the pellet containing the cell walls was further purified by sequential incubation and centrifugation in cold Tris-HCl, ethanol, acetone, a mixture of methanol: chloroform, and again acetone and ethanol. Cell wall extracts were treated with pronase in phosphate (pH 7.0). The walls were further treated with CDTA, 1 and 4 M KOH for pectin, hemicellulose I, and II extraction. Residual insoluble sediments were designated as the ‘cellulose fraction’. The amount of total sugar in each fraction was measured using the phenol-sulphuric acid method (Dubois et al. 1951) and the meta-hydroxy diphenyl method (Blumenkrantz and Asboe-Hansen 1973).

Statistical analysis

All data were analysed using an ANOVA and the means were compared using Duncan's multiple range test at $P < 0.05$. Correlations among the compositions, extension distances, elastic parameters, and plastic parameters of the root-cell-wall and root growth were analysed by Pearson’s correlations at $P < 0.05$. SPSS 21 software (IBM SPSS, USA) was used for all statistical analyses.

Results

Root growth

Salinity severely depressed root growth in all cultivars (Image 1, Table 1). The relative root growth in the sensitive cultivars (Yongliang-15, GS-6058) was lower than that of the tolerant cultivars (JS-7, Xinchun-31). Compared with the control, the roots became thicker under NaCl treatment, i.e., root diameters increased by ~10% and 40%, and the area of the root cross sections increased by ~1.2 and ~1.9 times in the sensitive and tolerant cultivars, respectively.

Extension of the root-cell-wall

The results of the elastic parameters and viscosity coefficients are shown in Table 2, wherein an increase in the E_0 value indicates a decrease in elasticity. The E_0 values in the sensitive cultivars were significantly increased after the 80 mM NaCl treatment, i.e. to almost double the values observed under the control treatment, However, no significant changes were observed in the tolerant cultivars after NaCl treatment. Under the non-saline condition, the E_0 values of the sensitive cultivars were significantly lower compared with those of the tolerant cultivars. The elastic modules of E_1, E_2, and E_3 approximately ranged from 2.78×10^7 to 4.87×10^7 Pa in all treatments, i.e. were ~10 times higher than E_0. No significant differences were observed among the E_1, E_2, and

E_3 modules between the 0 and 80 mM NaCl conditions for all cultivars, and no significant differences were detected in the viscosity modules (η_0, η_1, η_2, η_3) across cultivars and treatments.

Typical root extension curves of all cultivars under both 0 and 80 mM NaCl treatments were successfully obtained using the setting conditions. Representative extension curves are shown in Fig. 1. As expected, salinity treatment depressed the root extension and this depression was much more prominent in the salt-sensitive cultivars than in the tolerant cultivars. Extension and viscoelastic parameters are simply illustrated in Fig. 1. Further details regarding the extension curves have been described by Tanimoto et al. (2000). The elastic, plastic, and total extensions are shown in Fig. 2. The directly measured total extension distances of roots were decreased by about 40–60% after 80 mM NaCl treatment in all cultivars (Fig. 2A). The elastic and plastic extension distances were generally decreased by the salinity treatment in all cultivars (Fig. 2A). The converted extension distances, which eliminated the effect of root thickness on the salinity treatment, were all increased in the four cultivars because the NaCl treatment caused the roots to thicken (Fig. 2B). However, a significant decrease in the elastic and total extension in the sensitive cultivars was still observed after treatment with 80 mM NaCl, compared with the control. While no significant differences were detected in the elastic, plastic, and total extension results between the 0 and 80 mM NaCl treatments in the tolerant cultivars, elastic extension accounted for approximately one-half to two-thirds of the total extension in all cultivars and treatments, and plastic extension accounted for half or less than half of the total extension.

Chemical composition of the root-cell-wall

The chemical compositions and their relative amounts are shown in Fig. 3. The relative contents of the cell wall compositions were consistent with their absolute values. Irrespective of the wheat cultivars, no significant differences were detected in the total amounts of the root-cell-wall in the 10 mm-long apical root segments between the 0 and 80 mM NaCl treatments. However, the relative content of the four compositions (pectin, hemicellulose I, hemicellulose II, and cellulose) differed greatly in response to the NaCl treatment. The relative pectin content decreased, whereas the relative cellulose content increased in all cultivars under the saline condition. The tolerant cultivars showed significantly low relative hemicellulose I contents compared with the sensitive cultivars. The sensitive cultivars showed no significant changes in the relative hemicellulose II contents but the tolerant cultivars showed a significant decrease under the saline condition. Notably, the total cell wall content in Xinchun-31 was only about half that of the other cultivars. This may be due to differences in genetic background of this cultivar. The Xinchun-31 is a Chinese-Mexican hybird, while the other three cultivars are of Chinese origin.

This may be due to the different genetic background of this cultivar. The maternal origin of Xinchun-31 is Mexican, while the other three cultivars are Chinese origins.

Correlations among the root extension parameters and the cell wall compositions are shown in Fig. 4. It is noteworthy that negative correlations were detected between E_0 and relative pectin, E_0 and relative hemicellulose I, relative pectin and relative cellulose, and relative hemicellulose I and η_0; and positive correlations were detected between the root growth, the total and plastic extensions, and the relative pectin contents. In addition, when using the calculated extensions, a positive correlation was observed between the root growth and elastic extension.

Discussion

The tolerant cultivars (JS-7, Xinchun-31) showed higher relative root growth than the sensitive cultivars (Yongliang-15, GS-6058), which is consistent with their growth and production in real saline soils. Many previous reports have shown that the whole wheat growth is consistent with root growth under saline conditions (Sadat Noori and McNeilly 2000; Aslan et al. 2016; Mujeeb-Kazi et al. 2019). Therefore, we suggest that relative root growth at the early seedling stage can be used as a reliable salinity tolerance parameter for wheat cultivars. Hereafter, discussions regarding the salinity tolerance are based on the observed root growth.

Extension curve of the root-cell-wall

Extensibility of the cell wall is an important factor that regulates cell elongation in plant tissue (Sakurai 1991; Cosgrove 2018). Extension curves of the root-cell-wall (subjected to measurement using a creep meter) have only previously been reported for green peas, i.e. the first attempt using a creep meter to obtain the extension curve of the root-cell-wall (Tanimoto et al. 2000). The extension curves of wheat roots under both saline and non-saline conditions in this study showed similar shapes to that reported for green peas (Fig. 1). This result confirmed that the mechanical properties of plant roots, even thin wheat roots, follow the Kelvin-Voigt-Burgers viscoelastic model (Tanimoto et al. 2000). In the present study, the extension curves intuitively illustrated the cultivar differences in root-cell-wall extension and the effects of salinity on cell wall extension. The largely depressed extension in the sensitive cultivars indicated that the mechanical properties of the root-cell-wall of these cultivars were very sensitive to salinity stress.

Elastic parameter E_0 in relation to root growth

E_0 values have been reported for only three plants, i.e. green pea (Tanimoto et al.

2000), sorghum (Hattori et al. 2003), and Arabiadopsis (Shigeyama et al. 2016). In the present study, the E_0 of the elongation region of salt-sensitive cultivars under the non-saline condition ranged from 1.6 to 1.8 10^6 Pa (Table 2). These values were similar to those reported for green pea and sorghum roots (1.6–2.6 10^6 Pa) but were 10 times those reported for Arabiadopsis stems (1.8–3.2 10^5 Pa). The E_0 of the tolerant cultivars was higher than that of the above-mentioned plants, i.e. ~3.5 10^6 Pa. Salinity increased the E_0 in the sensitive cultivars but had no effect on that in the tolerant cultivars (Table 2). These results are very similar to those seen for Al stress, e.g. Al increased the E_0 in Al-sensitive wheat cultivar but had no effect on the E_0 in the tolerant cultivar (Ma et al. 2004). This previous report suggested that Al binding with the cell wall resulted in the deformation of the cell wall, which increased the E_0 but reduced the exentsibility. Sodium (Na^+) also directly binds with cell walls, and, the ion-binding was reportedly much lower in tolerant cultivars compared with sensitive cultivars in barley and *Silene paradoxa* (Flowers and Hajibagheri 2001; Colzi et al. 2012). Therefore, the increased E_0 values in the sensitive cultivars may have been partially due to excesive Na-binding with the cell walls, although the Na binding in the tolerant cultivars may have been insufficient to cause cell wall deformation. In the present study, the significantly increased E_0 values in the sensitive cultivars (Table 2) may represent one of the factors that inhibited the root growth. In contrast, the unaffected E_0 of the tolerant cultivars suggested that this parameter may not be a limiting factor for root growth in these studied cultivars under saline conditions. In addition, these results suggested that the mechanical properties of the root-cell-wall may be related to the cultivar differences in root growth under salinity stress. The turgor pressure of cells, i.e. the driving force for cell elongating, decreases under salinity stress (Rygol and Zimmermann 1990; Ogawa and Yamauchi 2006); therefore, the significance of cell wall elasticity on cell elongation becomes very pronounced under saline conditions.

Root growth has been found to be associated with the extensibility of root-cell-wall under drought (Hattori et al., 2003; Fan et al., 2006) and Al stress (Ma et al., 2004; Safari et al. 2018) conditions. Collectively, these previous findings and the findings of the present study indicate that the maintenance of the root-cell-wall extensibility is important for root growth under abiotic stress conditions. Our findings revealed that the E_0 was only about 1/10 of that of E_{1-3} and almost no significant differences were detected among E_{1-3} across treatments and cultivars (Table 2).

Cell wall extension and viscosity coefficient η in response to salinity treatment

Different elasticity traits of the cultivars resulted in different elastic and total extension distance in this study (Fig. 2A). After accounting for the changes in root thickness (which increased under salinity, Table 1), the extension distances of the tolerant cultivars under the saline condition were almost consistent with those under the control conditions (Fig. 2B). These findings suggested that the wall elastic property of the tolerant cultivars favoured cell elongation under the saline condition. The elastic extension accounted for approximately one-half to two-thirds of the total extension (Fig. 2), thus indicating that the elasticity of the cell wall mostly contributes to the cell extension in wheat. On the other hand, plastic extension accounted for half or less than half of the total extension (Fig. 2B) and η_0, which represents the viscosity and determines the plastic extension, was not affected by the salinity in all cultivars (Table 2, Fig. 2B). Therefore, we showed that the elastic properties of the root-cell-wall are more prominent for root elongation than the plastic properties in wheat.

In the present study, the NaCl treatment had no effect on the viscosity (plastic property) of the root-cell-wall in all cultivars (Table 2), except for a slightly high value in Xinchun-31. Tanimoto et al. (2000) suggested that the decrease in viscosity is related to expansin and the removal of other proteins and calcium ions from the cell wall. Hattori et al. (2003) suggested that Si-hemicellulose and Si-pectin conjugates were responsible for the observed changes in root viscosity. Ma et al. (2004) suggested that interference in the binding of new wall materials with old materials increased the viscosity and decreased plastic extension. Shigeyama et al. (2016) reported that the accumulation of free xyloglucan oligosaccharides and the reduced molecular size of xyloglucan in hemicellulose can decrease the viscosity parameters. However, in the present study, the presence of Na^+ did not affect the viscosity coefficient (Table 2) and plastic extension in all cultivars (Fig. 2B). Since this property and other related plastic extension parameters (e.g. irreversible extension) are also important factors that affect cell elongation, further investigations are needed to clarify how wheat plants maintain this wall property under saline conditions.

Correlations between extension parameters and compositions

The comparable total cell wall amounts under the saline and non-saline conditions in all cultivars showed a stable allocation of carbon assimilation in the wheat cultivars, despite the growing environment (Fig. 3). The general decrease in the pectin and increase in the cellulose contents indicated a spatial-temporal change in cells under saline conditions.

To our knowledge, this is the first study to assess the numeric correlations among viscoelastic parameters and cell wall compositions. The negative correlations between E_0 and the relative pectin and hemicellulose I contents demonstrated the great contribution of these two compositions to cell elastic extension. Although the linkage of pectin-cellulose (Wang et al. 2015) and pectin-xylan (Tan et al. 2013) were reported, the load-bearing points are suggested to be hemicellulose II-cellulose conjunctions

(Cosgrove 2018). Therefore, higher amounts of pectin and hemicellulose I would be expected to benefit cell elongation. A high positive correlation between E_0 and η_0 reveals interactions between the elastic and viscosity properties of the cell wall. Previous reports, although not statistical supported, also showed a positive correlation between these two parameters (Hattori et al. 2003; Ma et al. 2004). These results imply that some wall constitutes contribute to both elastic and viscos properties of cell wall.

The negative correlation between the relative pectin and cellulose contents and the opposite correlations of these two compositions with the extension parameters (E_0, total, plastic, and elastic extension, Fig. 4) indicate that the deposition of cellulose to the growing cell wall restricts the elongation of the cell while higher amounts of pectin improves cell elongation. This notion is consistent with the report by An et al. (2014b), who showed that an increase in the pectin content induced by Ca application enhanced root growth in soybean. Contrasting effects of pectin and cellulose on cell wall extension have been reported for white sprunce (Renault and Zwiazek 1997). In the present study, the final root growth under the saline condition was determined to be positively correlated with the total, elastic, and plastic extensions, as well as the relative pectin content (Fig. 4). These results revealed the significance of the root-cell-wall properties and the special role that pectin plays in root growth under salinity stress.

Based on the growth processes and dynamics of the cell wall (Cosgrove 2018), our results implied that the loosening of root cell wall under saline conditions (with reduced turgor pressure) was largely depressed in the sensitive cultivars but maintained to some extent in the tolerant cultivars. This wall loossenning corresponded to the elastic extension. When the root-cell-wall loosens, new wall materials fill in the space or bind to the old wall. These materials improve the viscosity and their levels correspond with the plastic nature, i.e. the final elongation, of the root region. Present study revealed the regulation role of cell wall in root growth. Cultivar difference in salt tolerance may be related with the property of root cell wall.

In conclusion, 1) Salinity decreased the root-cell-wall extension in salt-sensitive cvs through an increased E0. However, there were no significant effects on the salt-tolerant cultivars; 2) The elastic properties of root-cell-wall of wheat under salinity were more pronounced in root elongation as compared with the plastic properties; 3) Increment in pectin and hemicellulose-I better improved the elastic extension in the root-cell-wall, relative to the deposition of cellulose. Further studies on the changes, constitutions, and functions of the chemical compositions with regards to the cell wall extension in various crops are needed to fully understand the role of cell walls in root growth under abiotic stresses.

Declarations

Funding: This study is partially funded by the Grant-in-Aid for Scientific Research (C) of Japan Society for the Promotion of Science, No. 26450020.

Conflicts of Interest: The authors declare that they have no conflict of interest.

Ethics approval

Not applicable

Consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and material

All data generated or analysed during this study are included in this published article

Code availability

Not applicable

Authors' contributions

Yang Shao designed and carried out the experiments, analyzed the results. Yang Shao and Ping An wrote the manuscript. Xiaohui Feng, Irshad Muhammad, Victoria Otie, Weiqiang Li and Yuanrun Zheng provided scientific advice, and revised the manuscript. Ping An conceived the research area and supervised the project.

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Tables

Table 1 Root growth, diameter, and cross-sectional area of four wheat cultivars under 0 and 80 mM NaCl treatments

Cultivars	NaCl (mM)	Root length (cm)	Relative root growth (%)	Root diameter (mm)	Area of cross section (mm^2)	Increase in the cross-sectional area (%)
Yongliang-15	0	14.10 ± 0.58 a	100	0.524 ± 0.027 cd	0.216 ± 0.014 d	0
	80	5.15 ± 0.08 f	37	0.590 ± 0.005 b	0.273 ± 0.005 b	23
GS-6058	0	13.29 ± 0.92 a	100	0.531 ± 0.002 c	0.223 ± 0.003 cd	0
	80	3.77 ± 0.19 f	28	0.581 ± 0.005 b	0.264 ± 0.005 bc	18
JS-7	0	13.80 ± 0.14 a	100	0.426 ± 0.003 e	0.143 ± 0.002 e	0
	80	6.59 ± 0.33 e	49	0.590 ± 0.023 b	0.274 ± 0.023 b	93
Xinchun-31	0	15.84 ± 0.27 a	100	0.484 ± 0.001 d	0.184 ± 0.003 de	0
	80	8.98 ± 0.13 d	57	0.637 ± 0.027 a	0.320 ± 0.028 a	78

Values of root length, diameter and area of cross section represent means ± SEs (n = 4)

Table 2 Distribution of elastic parameters and viscosity coefficients of root cell walls of four wheat cultivars under 0 and 80 mM NaCl treatments

Cultivars	NaCl (mM)	Elastic parameters				Viscosity coefficients			
		E_0 (10^6 Pa)	E_1 (10^7 Pa)	E_2 (10^7 Pa)	E_3 (10^7 Pa)	η_0 (10^{10} Pa s)	η_1 (10^9 Pa s)	η_2 (10^8 Pa s)	η_3 (10^7 Pa s)
Yongliang-15	0	1.65 ± 0.21c	3.12 ± 0.25ab	3.31 ± 0.93a	3.60 ± 0.20a	2.07 ± 0.25b	1.30 ± 0.09a	2.03 ± 0.07a	4.33 ± 0.42a
	80	3.33 ± 0.15ab	3.00 ± 0.25b	3.09 ± 0.47a	4.15 ± 0.39a	2.58 ± 0.26b	1.21 ± 0.12a	1.76 ± 0.23a	5.12 ± 0.71a
GS-6058	0	1.95 ± 0.14bc	3.73 ± 0.19ab	3.40 ± 0.92a	4.64 ± 0.05a	2.69 ± 0.30b	1.56 ± 0.12a	2.02 ± 0.08a	4.89 ± 0.37a
	80	3.64 ± 0.44a	3.49 ± 0.35ab	3.51 ± 0.13a	4.19 ± 0.40a	1.83 ± 0.21b	1.37 ± 0.14a	1.81 ± 0.08a	4.76 ± 0.44a
JS-7	0	3.71 ± 0.49a	3.82 ± 0.14ab	3.13 ± 0.18a	3.83 ± 0.38a	2.86 ± 0.47b	1.68 ± 0.04a	2.03 ± 0.08a	4.34 ± 0.35a
	80	3.37 ± 0.50ab	3.41 ± 0.40ab	2.86 ± 0.29a	3.59 ± 0.39a	2.53 ± 0.30b	1.43 ± 0.14a	1.66 ± 0.16a	4.23 ± 0.58a
Xinchun-31	0	3.40 ± 0.29ab	4.34 ± 0.69a	3.35 ± 0.81a	3.64 ± 0.92a	3.55 ± 0.65ab	1.72 ± 0.32a	1.85 ± 0.26a	4.52 ± 1.00a
	80	4.04 ± 0.91a	4.28 ± 0.51ab	3.98 ± 0.11a	4.89 ± 1.57a	5.31 ± 1.66a	1.60 ± 0.10a	1.79 ± 0.35a	3.75 ± 1.28a

Values represent means ± SEs (n = 17–51)

Means followed by the same letter in the same column are not significantly different ($P < 0.05$)

Figure Legends

Fig. 1 Typical creep extension curves of root cell walls during the extension (5 min) and shrinkage (5 min) of four wheat cultivars under 0 (black lines) and 80 (grey lines) mM NaCl treatments. The total, elastic, and plastic extensions were determined by reading the extensions at 5 and 10 min. The schematic illustration is based on the extension curve of Yongliang-15. A Kelvin-Voigt-Burgers model with four elastic (E0, E1, E2, E3) and four plastic (η0, η1, η2, η3) parameters effectively analysed cell wall extension and shrinkage in the creep-extension analysis (Tanimoto et al. 2000). E and η describe the resistances of the material to the stretch force (P_0) in elastic and plastic extending process, respectively. Footnotes of 0-3 indicate the different extension stages.

P_0/E_0: linear instantaneous deformation

P_0/E_1+ P_0/E_2+ P_0/E_3: non-linear deformation

P_0/η_0: creep deformation

Fig. 2 The elastic, plastic, and total extensions of the root cell wall of four wheat cultivars under 0 and 80 mM NaCl treatments. A: Data were directly measured using a creep meter. B: Converted data that account for changes in root thickness. The converted extension = measured extension distance $\times$(1+ (S_{80}-S_0)/S_0). Where S_{80} and S_0 are areas of root cross section under 80 and 0 mM NaCl. Data represent means ± SEs (n = 17–51). Different upper- and lowercase letters indicate significant differences ($P < 0.05$) in the elastic extension and plastic extension, respectively

Fig. 3 Relative and absolute contents of pectin, hemicellulose I, hemicellulose II, and cellulose in the root cell wall of four wheat cultivars under 0 and 80 mM NaCl treatments. Values inside the bars indicate the relative values. Data represent means ± SEs (*n* = 5). Different letters within the same composition indicate significant differences in the relative content ($P < 0.05$). ns: no significant difference in the total cell wall content between 0 and 80 mM NaCl treatments within the same cultivar

Fig. 4 Heat map showing the correlations of extension, viscoelastic parameters, and composition of the root cell wall and root growth in wheats. HC I: Hemicellulose I; HC II: Hemicellulose II. E_0 and η_0 indicate the elastic and viscous properties of the root-cell-wall, respectively. Red and blue colour indicate positive and negative correlation, respectively. Circle 大小？Significant correlations are indicated by asterisks (*P < 0.05, **P < 0.01). This graph is depicted by TBtools (Chen et al. 2020)削除？

Image 1 Roots of four wheat cultivars 10 days after 0 and 80 mM NaCl treatments.