ISSN 1392-3196 / e-ISSN 2335-8947
Zemdirbyste-Agriculture, vol. 109, No. 1 (2022), p. 55–62
DOI 10.13080/z-a.2022.109.008

Iron deficiency impacts chlorophyll biosynthesis, leaf cell expansion, xylem development and physiology of *Prunus persica* grafted onto rootstocks Garnem and GF 677

Servet ARAS, Hakan KELES, Erhan BOZKURT

Yozgat Bozok University, Faculty of Agriculture
66200 Yozgat, Turkey
E-mail: servet.aras@yobu.edu.tr

Abstract

Rootstocks tolerant to iron (Fe) deficiency can be used to cope with Fe chlorosis damage. In the experiment, two peach (*Prunus persica* (L.) Batsch) rootstocks Garnem and GF 677 grown under Fe deficiency conditions were compared. Plants were subjected to Fe deficiency for three months, and some leaf physiological and histological responses were assessed. The relative growth rate of scion diameter and the root to shoot dry weight ratio decreased in both rootstocks. Leaf malondialdehyde content increased in rootstocks Garnem and GF 677 by 22% and 15%, respectively. In leaves, total phenolic content decreased in both rootstocks. Leaf chlorophyll and chlorophyll precursor concentrations decreased under Fe deficiency. Midrib and xylem thickness, xylem conduit width and number of xylem conduits decreased because of Fe deficiency, and the decreases in the parameters were found higher in rootstocks Garnem. Iron triggered leaf cell division, but cell expansion could not occur due to the lack of Fe.
The results of the experiment demonstrated that Fe is a prerequisite for chlorophyll biosynthesis and leaf cell expansion, and GF 677 is a more tolerant rootstock to Fe deficiency compared to Garnem and can be used in peach orchards subjected to Fe deficiency conditions.

Keywords: cell division, cell expansion, iron, peach.

Introduction

Iron (Fe) is a prominent element for plants that has pivotal roles in plant growth, cell metabolism and nitrogen fixation (Eichert et al., 2010; Guo et al., 2020). Despite the presence of Fe in the soil, low solubility of Fe compounds due to the presence of carbonate, and high pH values in soils cause deficiency symptoms in plants (Aras et al., 2018). In leaves, Fe deficiency results in chlorosis due to a decline in chlorophyll synthesis (Guo et al., 2020). In many fruit trees, Fe chlorosis limits fruit yield and quality (Pestana et al., 2005; Valipour et al., 2020).

Peach (*Prunus persica* (L.) Batsch) is known to be sensitive to Fe deficiency (Molassiotis et al., 2006; Arıkan et al., 2018). The issue of Fe deficiency in peach orchards is a serious problem for the growers. Iron deficiency changes the physiology, biochemistry and anatomy of plants (Donnini et al., 2011). Alterations in gas exchange, chlorophyll fluorescence and antioxidants caused by Fe deficiency have been reported in peach (Molassiotis et al., 2006). Furthermore, oxidative stress occurred in quince rootstocks under Fe deprivation conditions (Valipour et al., 2020). Fernández et al. (2008) reported that Fe deficiency led to reduction in cuticular lipids and the size of stomatal guard cells in leaves of pear and peach. Stomatal functioning and xylem conductivity of leaves have been shown to be disturbed in peach under Fe deficiency (Eichert et al., 2010). The leaf size and area decreased in pear and peach under Fe deficiency (Fernández et al., 2008). Leaves are the energy factories of plants, and leaf growth occurs in two consecutive phases – leaf cell division and cell expansion (Gonzalez et al., 2012). The effect of nitrogen (MacAdam et al., 1989), phosphorus (Kavanová et al., 2006) and zinc (Jain et al., 2010) on leaf cell division and expansion has been studied, but the relationship between Fe and leaf cell division and expansion is still not completely known.

Peach orchards may be established in alkaline or calcareous soil, and Fe deficiency is a common issue for the growers (Arıkan et al., 2018). Thus, knowledge of the responses of peach rootstocks to Fe deficiency conditions is needed.

In the present study, the chlorophyll biosynthesis, histological and physiological responses to Fe deficiency in the peach cultivar 'Rich May' grafted onto rootstocks Garnem and GF 677 were assessed. Furthermore, the tolerance degree of Garnem and GF 677 to Fe deficiency was evaluated.

Please use the following format when citing the article:
Aras S., Keles H., Bozkurt E. 2022. Iron deficiency impacts chlorophyll biosynthesis, leaf cell expansion, xylem development and physiology of *Prunus persica* grafted onto rootstocks Garnem and GF 677. Zemdirbyste-Agriculture, 109 (1): 55–62. DOI 10.13080/z-a.2022.109.008

Materials and methods

The experiment was performed in March of 2020 in a climate-controlled greenhouse at the Yozgat Bozok University, Turkey. The peach cultivar 'Rich May' scions on rootstocks GF 677 (*Prunus dulcis* × *P. persica*) and Garnem (*P. dulcis* × *P. persica*) were used. The saplings were cultivated in 10 L pots containing perlite. The perlite was used as growing media due to deficiency of nutrients. The experiment was laid out in a randomized plot design with 3 replications, with 5 plants per replication. All plants were treated with Hoagland's nutrient solution of the following composition: 5 mM $Ca(NO_3)_2$, 5 mM KNO_3, 2 mM $MgSO_4$, 1 mM KH_2PO_4, 25 µM H_3BO_3, 2 µM $MnSO_4$, 2 µM $ZnSO_4$, 0.5 µM $CuSO_4$, 0.4 µM $(NH_4)_6Mo_7O_{24}$ and 20 µM Fe-EDDHA (Fe-ethylenediamine-di-o-hydroxy-phenylacetic acid) (Hoagland, Arnon, 1950) for two months, and for iron (Fe) fertilization, Fe-EDDHA source was used. Then, plants were exposed to Fe-deficient Hoagland's solution (except for the control) for 3 months, and control plants were watered with the Hoagland's solution.

Morphological measurements. The relative growth rate (RGR) of the scion diameter was calculated according to Del Amor and Marcelis (2003). Dry weight (DW) of root to shoot ratio was calculated after drying the root and shoot parts in an oven at 72°C temperature for 48 h.

Physiological measurements. Lipid peroxidation was determined by estimating the leaf malondialdehyde (MDA) content according to Madhava Rao and Sresty (2000). Protein levels were estimated by the method of Bradford (1976), as the standard using bovine serum albumin. The leaf proline content was estimated by the method of Bates et al. (1973). Chlorophyll *a*, *b*, *a* + *b* content and carotenoids concentration were analysed according to Corte-Real et al. (2017). A total of 0.5 g leaf sample was ground in 10 ml extract (80% acetone) and stored in the dark at 25°C temperature for 24 h. Before spectrophotometer measurements the sample was filtered. Absorbance was measured at 470, 645 and 663 nm. For determination of total anthocyanin content, fresh leaves (1 g) of three plants per replicate were homogenized in methanol containing 1% HCl (v/v) at 4°C temperature and incubated for 24 h. After centrifugation, the absorbance of the supernatant was measured at 530 and 657 nm, and total anthocyanin content was calculated as in Mita et al. (1997). Total phenolic content was evaluated by the Singleton and Rossi (1965) method.

Chlorophyll precursors. The concentrations of protoporphyrin IX (Proto IX), Mg-protoporphyrin IX (Mg-Proto IX) and protochlorophyllide (Pchlide) were determined as described by Hodgins and Van Huystee (1986) and Liu et al. (2015). Chlorophyll yield was estimated by chlorophyll *a* + *b*/Proto IX (Aras et al., 2021).

Tolerance indices to Fe deficiency according to alterations in RGR, MDA and chlorophyll content were evaluated:

tolerance indices of RGR of scion diameter = (RGR of scion diameter of Fe-deficient plant / RGR of scion diameter of control) × 100;

tolerance indices of MDA = (MDA of Fe-deficient plant / MDA of control) × 100;

chlorophyll stability index = (chlorophyll *a* + *b* of Fe-deficient plant / chlorophyll *a* + *b* of control) × 100 (Shetty et al., 1995; Aras et al., 2020).

Histological studies. The midrib of the leaf tissue of three plants per replicate was observed. Toluidine Blue O (O'Brien et al., 1964) and acid phloroglucin dyes were used. The observations were done with a microscope Olympus CX21 coupled to a digital camera at 4× and 10× magnifications. The cortex, epidermis and xylem were measured. The number of the cortex cell layers was calculated from cortex thickness divided by cortical cell diameter.

Statistical analysis was carried out using software package *SPSS*, version 20.0 (IBM Inc., USA). Data were subjected to two-way analysis of variance (ANOVA) and were separated by the Duncan's test at a significance level of $P < 0.05$.

Results

Visual symptoms of leaf chlorosis were observed in young leaves under Fe deficiency conditions. The chlorosis was observed in the interveinal area. The symptoms were severe in rootstock Garnem compared to GF 677 (Figure 1).

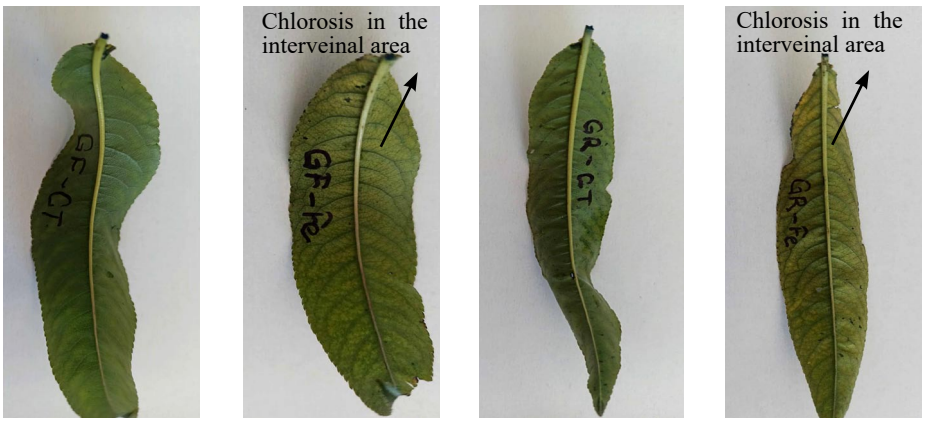

Figure 1. Visual symptoms of Fe deficiency in peach leaves

Peach plant morphology was significantly affected by Fe deficiency (Table 1). RGR of scion diameter and root to shoot dry weight (DW) ratio decreased in both rootstocks. The decrease in both parameters was larger in rootstock Garnem compared to GF 677.

Plant physiological parameters were influenced by Fe deficiency. Under Fe deficiency, leaf MDA concentration increased in Rich May/Garnem and Rich May/GF 677 by 22% and 15%, respectively (Table 2). Protein concentration increased in both rootstocks, whereas that of proline decreased under Fe deficiency conditions. Under Fe deficiency, carotenoids concentration increased in peach plants, and a further increment was found in GF 677 (Table 3). Anthocyanin

Table 1. Effects of Fe deficiency on relative growth rate (RGR) and root to shoot ratio of peach plants

Cultivar / rootstock	Treatment	RGR of scion diameter	Root to shoot ratio in DW
Rich May/Garnem	control	0.531 a	0.521 a
	Fe deficiency	0.301 b	0.344 b
Rich May/GF 677	control	0.333 a	0.675 a
	Fe deficiency	0.272 b	0.545 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

Table 2. Effects of Fe deficiency on malondialdehyde (MDA) and protein and proline contents of peach plants

Cultivar / rootstock	Treatment	MDA μmol g^{-1} FW	Protein μg g^{-1} FW	Proline μmol g^{-1} FW
Rich May/Garnem	control	0.00225 b	0.0414 b	1.79 a
	Fe deficiency	0.00275 a	0.0493 a	1.37 b
Rich May/GF 677	control	0.00241 b	0.0369 b	1.46 a
	Fe deficiency	0.00276 a	0.0496 a	0.88 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

Table 3. Effects of Fe deficiency on carotenoids concentration and anthocyanin and total phenolic contents of peach leaves

Cultivar / rootstock	Treatment	Carotenoids mg L^{-1}	Anthocyanins mg L^{-1}	Total phenolics μg GAE 100 g^{-1} FW
Rich May/Garnem	control	0.33 b	7.29 b	2.22 a
	Fe deficiency	5.34 a	12.75 a	1.79 b
Rich May/GF 677	control	1.00 b	7.58 a	2.20 a
	Fe deficiency	4.82 a	6.39 b	1.78 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$; GAE – gallic acid equivalent.

concentration increased in Rich May/Garnem, while it decreased in Rich May/GF 677. Under Fe deficiency, total phenolic content decreased in Rich May/Garnem and Rich May/GF 677 by 24% and 19%, respectively.

Chlorophyll content decreased under Fe deficiency (Table 4). Further declines in chlorophyll *a*, *b*, *a* + *b* and *a* to *b* ratio values were found in rootstock Garnem compared to GF 677. Iron deficiency also decreased the concentration of the chlorophyll precursors tested (Table 5). Similar to the chlorophyll content, Fe deficiency in rootstock Garnem had higher declines in the concentration of the precursors.

Table 4. Effects of Fe deficiency on chlorophyll *a*, *b*, *a* + *b* and *a* to *b* ratio of peach leaves

Cultivar / rootstock	Treatment	Chlorophyll *a* μg g^{-1} FW	Chlorophyll *b* μg g^{-1} FW	Chlorophyll *a* + *b* μg g^{-1} FW	Chlorophyll *a* to *b* ratio
Rich May/Garnem	control	35.31 a	52.08 a	87.26 a	1.47 a
	Fe deficiency	23.57 b	12.74 b	36.21 b	0.54 b
Rich May/GF 677	control	34.20 a	46.38 a	80.39 a	1.35 a
	Fe deficiency	32.73 b	23.09 b	55.55 b	0.70 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

Table 5. Effects of Fe deficiency on chlorophyll precursors of peach leaves

Cultivar / rootstock	Treatment	Proto IX μg g^{-1} FW	Mg-Proto IX μg g^{-1} FW	Pchlide μg g^{-1} FW
Rich May/Garnem	control	0.1034 a	0.0601 a	0.0401 a
	Fe deficiency	0.0750 b	0.0329 b	0.0170 b
Rich May/GF 677	control	0.1524 a	0.0722 a	0.0376 a
	Fe deficiency	0.1002 b	0.0448 b	0.0235 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

To compare Fe deficiency tolerance of the rootstocks, tolerance indices were evaluated (Table 6). Under Fe deficiency, tolerance indices of RGR of scion diameter and chlorophyll stability index (CSI) were higher in Rich May/GF 677 compared to Rich May/Garnem. Tolerance indices of MDA content were higher in Rich May/Garnem, which demonstrates that cell damage was higher in Rich May/Garnem.

Table 6. Effects of Fe deficiency on tolerance indices (TI) of relative growth rate (RGR), malondialdehyde (MDA) content and chlorophyll stability index (CSI) of peach plants

Cultivar / rootstock	TI of RGR of scion diameter	TI of MDA	CSI
Rich May/Garnem	56.64 b	122 a	41.49 b
Rich May/GF 677	81.56 a	114 b	69.09 a

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

Histological responses were significantly affected by Fe deficiency. Midrib anatomy including cortex, epidermis and xylem was evaluated (Figure 2). Toluidine blue dye stained cortical cells as blue (Figure 2A, B) and phloroglucinol stained xylem conduits as red (Figure 2C).

The cortical cell diameter and cortex thickness decreased in both rootstocks under Fe deficiency, while the number of cortex cell layers increased (Table 7).

These results demonstrate that Fe triggered leaf cell division, but cell expansion did not occur due to the lack of Fe. Epidermis thickness also decreased under Fe deficiency. Midrib and xylem thickness, xylem conduit width and number of xylem conduits were decreased by Fe deficiency, and the declines in the parameters were higher in rootstock Garnem (Table 8).

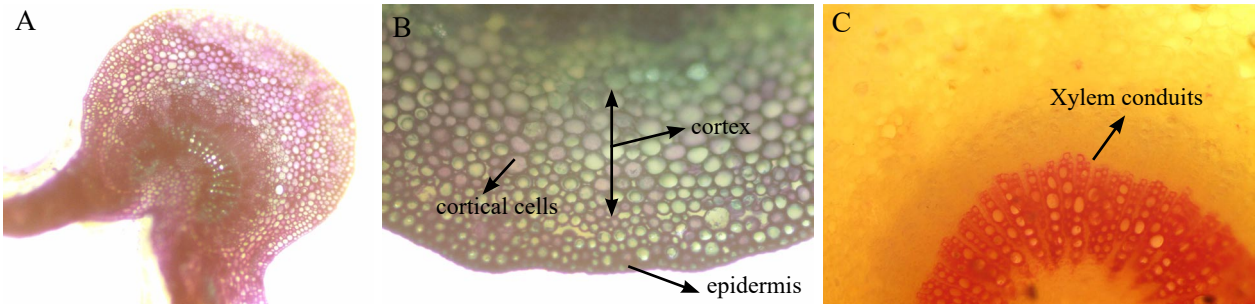

Figure 2. General view of cross-sectional images of midrib, stained by Toluidine blue dye (A), cortex cells at 10× magnification (B) and xylem of midrib stained by phloroglucinol (C)

Table 7. Effects of Fe deficiency on cortical cell diameter, cortex thickness, number of cortex cell layers, epidermis thickness and lamina thickness of peach leaf midribs

Cultivar / rootstock	Treatment	Cortical cell diameter µm	Cortex thickness µm	Number of cortex cell layers	Epidermis thickness µm	Lamina thickness µm
Rich May/Garnem	control	38.95 a	384.6 a	9.87 b	18.4 a	167.3 a
	Fe deficiency	29.15 b	314.0 b	10.79 a	14.6 b	152.0 b
Rich May/GF 677	control	33.83 a	351.6 a	10.38 b	15.2 a	198.0 a
	Fe deficiency	27.55 b	306.0 b	11.11 a	12.7 b	173.6 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

Table 8. Effects of Fe deficiency on midrib and xylem thickness, xylem conduits width and number of xylem conduits of peach leaf midribs

Cultivar / rootstock	Treatment	Midrib thickness µm	Xylem thickness µm	Xylem conduits width µm	Number of xylem conduits 1000 µm^{-1}
Rich May/Garnem	control	1137 a	163.3 a	21.6 a	53.7 a
	Fe deficiency	943 b	150.6 b	19.7 b	48.5 b
Rich May/GF 677	control	752 a	126.0 a	17.8 ns	54.6 a
	Fe deficiency	671 b	118.3 b	17.4	50.4 b

Note. Means separation within column by Duncan's multiple range test at $P < 0.05$.

Discussion

Iron deficiency affected morphological, physiological and histological responses of peach cultivar 'Rich May' grafted onto rootstocks Garnem and GF 677. Many researchers (Eichert et al., 2010; Arıkan et al., 2018) have investigated the physiological responses of peach plants to Fe deficiency, but little is known about the chlorophyll biosynthesis and histology.

Morphological and physiological responses. Plant growth by determination of RGR of scion diameter and root to shoot ratio in dry weight and the growth parameters were depressed under Fe deficiency conditions. Under Fe deficiency, the decline was more pronounced in rootstock Garnem. Moreover, tolerance index of RGR of scion diameter was higher in Rich May/GF 677 compared to Rich May/Garnem. Under Fe deficiency conditions, shoot length decreased in citrus rootstocks (Pestana et al., 2005) and dry weight decreased of peach plantlets (Lombardi et al., 2003). To evaluate dry matter partitioning between shoots and roots, root to shoot ratio is used (Aras, Eşitken, 2019). Root to shoot ratio was decreased by Fe deficiency, and the further reduction was found in Rich May/Garnem compared to Rich May/GF 677.

Iron deficiency causes formation of reactive oxygen species (ROS) that increase damage in cell membranes (Kaya et al., 2019). Cell membrane damage was assessed by determination of lipid peroxidation. Iron deficiency caused increment in MDA a product of lipid peroxidation. Greater damage was found in rootstock Garnem compared to GF 677, which is shown as increment in tolerance index of MDA in Table 6. Similar results related to MDA increment by Fe deficiency were reported

in apple (Zhang et al., 2020) and quince (Valipour et al., 2020). Plants deal with stress factors by accumulation of some solutes and pigments including proteins, proline, carotenoids and anthocyanin (Reddy et al., 2004). Protein content and carotenoids concentration increased in Fe-deficient peach plants, and the increments were higher in rootstock GF 677 than in Garnem.

Proline is an amino acid, which plays prominent roles in stress tolerance (Szabados, Savouré, 2010). In our experiment, under Fe deficiency, proline content decreased in peach leaves. Arias-Baldrich et al. (2015) stated that proline is synthesized from glutamate (a nitrogen compound), and Fe deficiency exerts negative effects on nitrogen metabolism. Therefore, reduction in proline biosynthesis is an expected result under Fe deficiency conditions. Also, leaf phenolic content was evaluated. Phenolic compounds are able to increase Fe remobilization and absorption by plants (Valipour et al., 2020). Furthermore, phenolics take a pivotal role in lignin biosynthesis (Weng, Chapple, 2010). In the present experiment, under Fe deficiency, phenolic content of the peach leaves decreased.

Chlorophyll biosynthesis. Chlorophylls, the most abundant tetrapyrroles in plants, play a crucial role in photosynthesis (Pattanayak, Tripathy, 2011). Chlorophyll content decreased in *Prunus* species under many environmental stresses including mineral deficiency (Aras et al., 2021) and drought stress (Viljevac et al., 2013). In our experiment, under Fe deficiency conditions, chlorophyll *a*, *b* and *a* + *b* content decreased in both rootstocks. The reduction was higher in rootstock Garnem compared to GF 677, and chlorophyll stability index was higher in GF 677. Under Fe deficiency conditions, chlorophyll content was also declined in apple (Guo et al., 2020) and soybean (Santos et al., 2019).

Chlorophyll is a tetrapyrrole containing magnesium (Mg). The chlorophyll biosynthesis initiates with formation of 5-aminolevulinic acid (ALA) and continues with formation of other porphyrins including protoporphyrin IX (Proto IX), Mg-Proto IX and protochlorophyllide (Pchlide) (Tanaka, Tanaka, 2007). Leaf chlorosis may be a consequence of reduction in chlorophyll biosynthesis. Thus, under Fe deficiency, chlorophyll pathway was investigated to compare chlorosis degree of rootstocks Garnem and GF 677. The concentration of the chlorophyll precursors in peach leaves decreased under Fe deficiency, and the reduction was greater in rootstock Garnem.

In a previous experiment (Aras et al., 2021), the chlorophyll precursors in peach plants under calcium (Ca) deficiency conditions were evaluated. Proto IX was not significantly changed, and Ca deficiency hampered chlorophyll pathway at Mg-Proto IX step in rootstock Garnem. In the current experiment, Proto IX also remarkably decreased under Fe deficiency for both rootstocks. Reduction in the contents of the chlorophyll precursors were also reported under salinity (Wu et al., 2018) and osmotic (Niu, Ma, 2018) stress.

Histological responses. In higher plants, the conversion of young leaf to a mature leaf consists of two partially overlapping phases. In the first phase, cell division occurs; in the second phase, cell division has ceased and cell expansion initiates (Gonzalez et al., 2012). Thus, leaf development requires the success in both cell division and expansion. In the current experiment, cortical cells of leaf midrib were evaluated. Lead midrib is composed of specialized tissues (phloem and xylem) and other cells playing pivotal roles in water, mineral and solute transport in leaves (da Silva et al., 2015; Lechthaler et al., 2019). In cortical cell diameter and cortex thickness, Fe deficiency caused remarkable reduction; however, the number of cortex cell layers increased in both rootstocks. These results demonstrate that Fe deficiency increased cell division and decreased cell expansion in peach leaf midribs. Thus, Fe is involved in cell expansion. Under Fe deficiency conditions, the expansion phase was less inhibited and cell division was less increased in rootstock GF 677 compared to Garnem. Cell division and elongation decreased in grass leaves under P deficiency conditions (Kavanová et al., 2006).

In the experiment, leaves of Fe-deficient plants were brittle and dropped easily. It has been reported that cortex plays an important role in mineral acquisition (Malta et al., 2016), and it was considered that leaves increased cell division to uptake more Fe. However, leaf cell expansion did not occur under Fe deficiency, leaf cells required Fe to elongate and leaves easily dropped due to the lack of cell expansion. To the best of our knowledge, the current paper is the first report showing that Fe deficiency increased leaf cell division and inhibited cell expansion. Also, the effect of Fe deficiency on epidermis thickness of leaf midribs was studied; Fe deficiency decreased epidermis thickness in both rootstocks. Epidermis is an external barrier against environmental conditions (Onoda et al., 2015). Thus, Fe deficiency may make plants vulnerable to stress factors.

Xylem status of leaf midribs under Fe deficiency was evaluated. Xylem plays an important role in water and mineral transport from roots to shoots (Orłowska et al., 2013). In the experiment, xylem thickness, xylem conduit width and number decreased in both rootstocks under Fe deficiency conditions (Figure 2). Similar with our results, Eichert et al. (2010) found that Fe deficiency disturbed xylem development in peach cultivar 'Miraflores'. Xylem tissue is comprised of lignin (Whetten et al., 1998) and decline in some minerals leads reduction in lignification (Huang et al., 2019; Aras et al., 2021). Iron deficiency caused decrease in lignification and consequently reduction in xylem formation. Moreover, decreases in xylem, cortex and epidermis thickness led to the reduction in midrib thickness in both rootstocks.

Conclusion

Iron deficiency deleteriously affected plant growth, chlorophyll biosynthesis, cell membranes and leaf cell expansion in peach plants. Inhibition of cell expansion and decline in chlorophyll biosynthesis caused by Fe deficiency could explain the reduction in plant growth. Rootstock GF 677 was found to be more tolerant to Fe deficiency conditions compared to Garnem. Thus, rootstock GF 677 can be suggested to peach orchards under alkaline or calcareous soil conditions.

Acknowledgements

This study was supported by the grants from Yozgat Bozok University with Project Coordination Application and Research Center, No. 6602C-ZF/20-387.

Received 01 05 2021
Accepted 25 11 2021

References

Aras S., Eşitken A. 2019. Dry matter partitioning and salt tolerance via salicylic acid treatment in strawberry plant under salt stress. KSU Journal of Agriculture and Nature, 22: 337–341. https://doi.org/ 10.18016/ksutarimdoga.vi.545825

Aras S., Arıkan Ş., İpek M., Eşitken A., Pırlak L., Dönmez M. F., Turan M. 2018. Plant growth promoting rhizobacteria enhanced leaf organic acids, FC-R activity and Fe nutrition of apple under lime soil conditions. Acta Physiologiae Plantarum, 40 (6): 120. https://doi.org/10.1007/s11738-018-2693-9

Aras S., Keles H., Eşitken A. 2020. SNP mitigates malignant salt effects on apple plants. Erwerbs-Obstbau 62 (1): 107–115. https://doi.org/10.1007/s10341-019-00445-1

Aras S., Keles H., Bozkurt E. 2021. Physiological and histological responses of peach plants grafted onto different rootstocks under calcium deficiency conditions. Scientia Horticulturae, 281: 109967. https://doi.org/10.1016/j.scienta.2021.109967

Arias-Baldrich C., Bosch N., Begines D., Feria A. B., Monreal J. A., García-Mauriño S. 2015. Proline synthesis in barley under iron deficiency and salinity. Journal of Plant Physiology, 183: 121–129. https://doi.org/10.1016/j.jplph.2015.05.016

Arıkan Ş., Eşitken A., İpek M., Aras S., Şahin M., Pırlak L., Dönmez M. F., Turan M. 2018. Effect of plant growth promoting rhizobacteria on Fe acquisition in peach (*Prunus persica* L.) under calcareous soil conditions Journal of Plant Nutrition, 41 (17): 2141–2150. https://doi.org/10.1080/01904167.2018.1482910

Bates L. S., Waldren R. P., Teare, I. D. 1973. Rapid determination of free proline for water-stress studies. Plant and Soil, 39: 205–207. https://doi.org/10.1007/BF00018060

Bradford M. M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry, 72: 248–254. https://doi.org/10.1016/0003-2697(76)90527-3

Corte-Real J., Bertucci M., Soukoulis C., Desmarchelier C., Borel P., Richling E., Hoffmann L., Bohn T. 2017. Negative effects of divalent mineral cations on the bioaccessibility of carotenoids from plant food matrices and related physical properties of gastro-intestinal fluids. Food and Function, 8 (3): 1008–1019. https://doi.org/10.1039/C6FO01708H

da Silva N. R., Florindo J. B., Gómez M. C., Rossatto D. R., Kolb R. M., Bruno O. M. 2015. Plant identification based on leaf midrib cross-section images using fractal descriptors. PLoS ONE, 10 (6): e0130014. https://doi.org/10.1371/journal.pone.0130014

Del Amor F., Marcelis L. F. M. 2003. Regulation of nutrient uptake, water uptake and growth under calcium starvation and recovery. Journal of Horticultural Science and Biotechnology, 7: 343–349. https://doi.org/10.1080/14620316.2003.11511629

Donnini S., Dell'Orto M., Zocchi G. 2011. Oxidative stress responses and root lignification induced by Fe deficiency conditions in pear and quince genotypes. Tree Physiology, 31 (1): 102–113. https://doi.org/10.1093/treephys/tpq105

Eichert T., Peguero-Pina J. J., Gil-Pelegrín E., Heredia A., Fernández V. 2010. Effects of iron chlorosis and iron resupply on leaf xylem architecture, water relations, gas exchange and stomatal performance of field-grown peach (*Prunus persica*). Physiologia Plantarum, 138 (1): 48–59. https://doi.org/10.1111/j.1399-3054.2009.01295.x

Fernández V., Eichert T., Del Río V., López-Casado G., Heredia-Guerrero J. A., Abadía A., Heredia A., Abadía J. 2008. Leaf structural changes associated with iron deficiency chlorosis in field-grown pear and peach: physiological implications. Plant and Soil, 311 (1): 161–172. https://doi.org/10.1007/s11104-008-9667-4

Gonzalez N., Vanhaeren H., Inzé D. 2012. Leaf size control: complex coordination of cell division and expansion. Trends in Plant Science, 17 (6): 332–340. https://doi.org/10.1016/j.tplants.2012.02.003

Guo A., Hu Y., Shi M., Wang H., Wu Y., Wang Y. 2020. Effects of iron deficiency and exogenous sucrose on the intermediates of chlorophyll biosynthesis in *Malus halliana*. PLoS ONE, 15 (5): e0232694. https://doi.org/10.1371/journal.pone.0232694

Hoagland D. R., Arnon D. I. 1950. The water-culture method for growing plants without soil. Circular 347. The College of Agriculture, University of California, 32 p.

Hodgins R. R., Van Huystee R. B. 1986. Rapid simultaneous estimation of protoporphyrin and Mg-porphyrins in higher plants. Journal of Plant Physiology, 125 (3–4): 311–323. https://doi.org/10.1016/S0176-1617(86)80153-5

Huang J. H., Xu J., Ye X., Luo T. Y., Ren L. H., Fan G. C., Qi Y. P., Li Q., Ferrarezi R. S., Chen L. S. 2019. Magnesium deficiency affects secondary lignification of the vascular system in *Citrus sinensis* seedlings. Trees, 33 (1): 171–182. https://doi.org/10.1007/s00468-018-1766-0

Jain R., Srivastava S., Solomon S., Shrivastava A. K., Chandra A. 2010. Impact of excess zinc on growth parameters, cell division, nutrient accumulation, photosynthetic pigments and oxidative stress of sugarcane (*Saccharum* spp.). Acta Physiologiae Plantarum, 32 (5): 979–986. https://doi.org/10.1007/s11738-010-0487-9

Kavanová M., Lattanzi F. A., Grimoldi A. A., Schnyder H. 2006. Phosphorus deficiency decreases cell division and elongation in grass leaves. Plant Physiology, 141 (2): 766–775. https://doi.org/10.1104/pp.106.079699

Kaya C., Akram N. A., Ashraf M. 2019. Influence of exogenously applied nitric oxide on strawberry (*Fragaria* × *ananassa*) plants grown under iron deficiency and/or saline stress. Physiologia Plantarum, 165 (2): 247–263. https://doi.org/10.1111/ppl.12818

Lechthaler S., Colangeli P., Gazzabin M., Anfodillo T. 2019. Axial anatomy of the leaf midrib provides new insights into the hydraulic architecture and cavitation patterns of Acer pseudoplatanus leaves. Journal of Experimental Botany, 70 (21): 6195–6201. https://doi.org/10.1093/jxb/erz347

Liu J., Wang J., Yao X., Zhang Y., Li J., Wang X., Xu Z., Chen W. 2015. Characterization and fine mapping of thermo-sensitive chlorophyll deficit mutant1 in rice (*Oryza sativa* L.). Breeding Science, 65: 161–169. https://doi.org/10.1270/jsbbs.65.161

Lombardi L., Sebastiani L., Vitagliano C. 2003. Physiological, biochemical, and molecular effects of *in vitro* induced iron deficiency in peach rootstock Mr. S 2/5. Journal of Plant Nutrition, 26 (10–11): 2149–2163. https://doi.org/10.1081/PLN-120024271

MacAdam J. W., Volenec J. J., Nelson C. J. 1989. Effects of nitrogen on mesophyll cell division and epidermal cell elongation in tall fescue leaf blades. Plant Physiology, 89 (2): 549–556. https://doi.org/10.1104/pp.89.2.549

Madhava Rao K. V., Sresty T. V. S. 2000. Antioxidative parameters in the seedlings of pigeonpea (*Cajanus cajan* L. Millspaugh) in response to Zn and Ni stress. Plant Science, 157: 113–128. https://doi.org/10.1016/S0168-9452(00)00273-9

Malta P. G., Arcanjo-Silva S., Ribeiro C., Campos N. V., Azevedo A. A. 2016. *Rudgea viburnoides* (Rubiaceae) overcomes the low soil fertility of the Brazilian Cerrado and hyperaccumulates aluminum in cell walls and chloroplasts. Plant and Soil, 408 (1): 369–384. https://doi.org/10.1007/s11104-016-2926-x

Mita S., Murano N., Akaike M., Nakamura K. 1997. Mutants of *Arabidopsis thaliana* with pleiotropic effects on the expression of the gene for β-amylase and on the accumulation of anthocyanin that are inducible by sugars. Plant Journal, 11: 841–851. https://doi.org/10.1046/j.1365-313X.1997.11040841.x

Molassiotis A., Tanou G., Diamantidis G., Patakas A., Therios I. 2006. Effects of 4-month Fe deficiency exposure on Fe reduction mechanism, photosynthetic gas exchange, chlorophyll fluorescence and antioxidant defense in two peach rootstocks differing in Fe deficiency tolerance. Journal of Plant Physiology, 163 (2): 176–185. https://doi.org/10.1016/j.jplph.2004.11.016

Niu K., Ma H. 2018. The positive effects of exogenous 5-aminolevulinic acid on the chlorophyll biosynthesis, photosystem and Calvin cycle of Kentucky bluegrass seedlings in response to osmotic stress. Environmental and Experimental Botany, 155: 260–271. https://doi.org/10.1016/j.envexpbot.2018.07.006

O'Brien, T., Feder, N., McCully, M. E. 1964. Polychromatic staining of plant cell walls by toluidine blue O. Protoplasma, 59 (2): 368–373. https://doi.org/10.1007/BF01248568

Onoda Y., Schieving F., Anten N. P. 2015. A novel method of measuring leaf epidermis and mesophyll stiffness shows the ubiquitous nature of the sandwich structure of leaf laminas in broad-leaved angiosperm species. Journal of Experimental Botany, 66 (9): 2487–2499. https://doi.org/10.1093/jxb/erv024

Orłowska E., Przybyłowicz W., Orlowski D., Mongwaketsi N. P., Turnau K., Mesjasz- Przybyłowicz J. 2013. Mycorrhizal colonization affects the elemental distribution in roots of Ni-hyperaccumulator *Berkheya coddii* Roessler. Environmental Pollution, 175: 100–109. https://doi.org/10.1016/j.envpol.2012.12.028

Pattanayak G. K., Tripathy B. C. 2011. Overexpression of protochlorophyllide oxidoreductase C regulates oxidative stress in Arabidopsis. PLoS ONE, 6 (10): e26532. https://doi.org/10.1371/journal.pone.0026532

Pestana M., de Varennes A., Abadía J., Faria E. A. 2005. Differential tolerance to iron deficiency of citrus rootstocks grown in nutrient solution. Scientia Horticulturae, 104 (1): 25–36. https://doi.org/10.1016/j.scienta.2004.07.007

Reddy A. R., Chaitanya K. V., Vivekanandan M. 2004. Drought-induced responses of photosynthesis and antioxidant metabolism in higher plants. Journal of Plant Physiology, 161 (11): 1189–1202. https://doi.org/10.1016/j.jplph.2004.01.013

Santos C. S., Ozgur R., Uzilday B., Turkan I., Roriz M., Rangel A. O., Carvalho S. M. P., Vasconcelos M. W. 2019. Understanding the role of the antioxidant system and the tetrapyrrole cycle in iron deficiency chlorosis. Plants, 8 (9): 348. https://doi.org/10.3390/plants8090348

Shetty K. G., Hetrick B. A. D., Schwab A. P. 1995. Effects of mycorrhizae and fertilizer amendments on zinc tolerance of plants. Environmental Pollution, 88 (3): 307–314. https://doi.org/10.1016/0269-7491(95)93444-5

Singleton V. L., Rossi J. R. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid. American Journal of Enology and Viticulture, 16: 144–158.

Szabados L., Savouré A. 2010. Proline: a multifunctional amino acid. Trends in Plant Science, 15 (2): 89–97. https://doi.org/10.1016/j.tplants.2009.11.009

Tanaka R., Tanaka A. 2007. Tetrapyrrole biosynthesis in higher plants. Annual Review of Plant Biology, 58: 321–346. https://doi.org/10.1146/annurev.arplant.57.032905.105448

Valipour M., Baninasab B., Khoshgoftarmanesh A. H., Gholami M. 2020. Oxidative stress and antioxidant responses to direct and bicarbonate-induced iron deficiency in two quince rootstocks. Scientia Horticulturae, 261: 108933. https://doi.org/10.1016/j.scienta.2019.108933

Viljevac M., Dugalić K., Mihaljević I., Šimić D., Sudar R., Jurković Z., Lepeduš H. 2013. Chlorophyll content, photosynthetic efficiency and genetic markers in two sour cherry (*Prunus cerasus* L.) genotypes under drought stress. Acta Botanica Croatica, 72 (2): 221–235. https://doi.org/10.2478/botcro-2013-0003

Weng J. K., Chapple C. 2010. The origin and evolution of lignin biosynthesis. New Phytologist, 187 (2): 273–285. https://doi.org/10.1111/j.1469-8137.2010.03327.x

Whetten R. W., MacKay J. J., Sederoff R. R. 1998. Recent advances in understanding lignin biosynthesis. Annual Review of Plant Biology, 49 (1): 585–609. https://doi.org/10.1146/annurev.arplant.49.1.585

Wu Y., Jin X., Liao W., Hu L., Dawuda M. M., Zhao X., Tang Z., Gong T., Yu J. 2018. 5-Aminolevulinic acid (ALA) alleviated salinity stress in cucumber seedlings by enhancing chlorophyll synthesis pathway. Frontiers in Plant Science, 9: 635. https://doi.org/10.3389/fpls.2018.00635

Zhang J. C., Wang X. F., Wang X. N., Wang F. P., Ji X. L., An J. P., Yang K., Zhao Q., You C. X., Hao Y. J. 2020. Abscisic acid alleviates iron deficiency by regulating iron distribution in roots and shoots of apple. Scientia Horticulturae, 262: 109018. https://doi.org/10.1016/j.scienta.2019.109018

Geležies trūkumas turi įtakos *Prunus persica* su Garnem ir GF 677 poskiepiais fiziologijai, chlorofilo biosintezei, lapų ląstelių dalijimuisi ir ksilemos raidai

S. Aras, H. Keles, E. Bozkurt

Yozgat Bozok universiteto Žemės ūkio fakultetas, Turkija

Santrauka

Siekiant sumažinti geležies (Fe) chlorozės žalą, gali būti panaudoti geležies trūkumui atsparūs poskiepiai. Eksperimento metu lyginti du paprastojo persiko (*Prunus persica* (L.) Batsch) poskiepiai Garnem ir GF 677, tris mėnesius auginti geležies trūkumo sąlygomis. Įvertintas augalo fiziologinis ir histologinis atsakas. Abiejų poskiepių ūglių skersmens santykinis augimo greitis ir šaknų bei ūglių sausosios masės santykis sumažėjo. Garnem ir GF 677 poskiepiuose lapų malondialdehido kiekis padidėjo atitinkamai 22 ir 15 %. Abiejų poskiepių lapuose sumažėjo bendras fenolinių junginių kiekis. Trūkstant geležies, sumažėjo lapų chlorofilo ir chlorofilo pirmtakų koncentracija. Dėl geležies trūkumo sumažėjo ksilemos storis ir kanalų plotis bei skaičius; šie rodikliai dar labiau sumažėjo Garnem poskiepio grupėje. Geležis paskatino lapų ląstelių dalijimąsi, tačiau dėl jos trūkumo ląstelės nedidėjo.

Eksperimento rezultatai parodė, kad geležis yra būtina chlorofilo biosintezės ir lapų ląstelių dalijimosi sąlyga, o poskiepis GF 677, palyginti su Garnem, yra atsparesnis geležies trūkumui ir gali būti naudojamas persikų soduose, kurių dirvožemiuose jos trūksta.

Reikšminiai žodžiai: geležis, ląstelių dalijimasis, ląstelių didėjimas, persikas.