

**TITLE: Changes induced by Fe deficiency and Fe resupply in the protein profile of GF 677 (*Prunus amygdalo x persica*) roots.**

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ABSTRACT: Peach is one of the fruit crops most affected by Fe chlorosis in the Mediterranean basin, and growers not using appropriate rootstocks and/or Fe fertilization face major losses in crop yield and quality. The aim of this work was to study changes induced by Fe deficiency in the root protein profile of the Fe chlorosis tolerant rootstock GF 677 (*Prunus amygdalo-persica*) grown in hydroponics. Also, root protein profile changes induced by Fe-resupply to Fe deficient plants were studied after 24 h of Fe application. Two-dimensional gel-based techniques were used to study the protein profiles of root extracts from plants grown in Fe sufficient, Fe-deficient and short-term Fe-resupply conditions, resolving  $338 \pm 5$ ,  $339 \pm 1$  and  $336 \pm 5$  spots, respectively, with 335 spots consistently detected and matched in all gels. Averaged maps for the three treatments were compared, using two batches of plants and in each of them three biological replicates pooling the roots from several plants. Iron deficiency caused above two-fold increases and  $>50\%$  decreases in relative abundance in 10 and 6 spots, respectively, whereas one spot was only detected in Fe-deficient plants. Iron resupply to Fe deficient plants, caused increases and decreases in relative abundance in 15 and 16 spots and one spot was only detected in Fe-resupplied Fe-deficient plants. Approximately 92% of the proteins changing in abundance were identified using nLC-ESI-MS/MS. Results show that Fe deficiency induced major changes in proteins related to oxidative stress (33%) and stress and defense responses (17%), followed by proteins related to protein synthesis and modification, energy and secondary metabolism (8% each). Proteins related to C and N metabolism accounted only for 8% of the changes. Iron resupplied plants presented a similar distribution of root protein profile changes when compared to that of Fe-sufficient plants. When comparing the root protein profile of Fe-resupplied plants to that of Fe-deficient plants, only 5 spots showed increases in relative abundance whereas 3 showed decreases. These results support that the effects of Fe fertilization on the root protein profile in the short term is limited.

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