Iron biofortification of oats (*Avena sativa cv. “Prevision”*) grown under aeroponic conditions

Luca Boschian (luca.boschian@phd.unipd.it)

TESAF (Dipartimento territorio e sistemi agroforestali), University of Padova, Padova, Italy

Tutor: Prof. Simone Vincenzi

The present contribution reports the results regarding the biofortification of oats aimed at increasing the final iron content of the whole plant. This plant, as many other monocotyledonous plants, absorbs iron through root-exudated compounds known as phytosiderophores. To enhance the iron absorption, plants were grown in iron-free nutritive solution and iron was later added as inorganic iron (iron (III) ammonium sulfate). This treatment was compared with standard nutrition with chelated iron (Fe-EDDHA). Different times of growth without iron and different harvesting post-iron addition were also investigated.

Biofortificazione del ferro in avena (*Avena sativa cv. “Prevision”*) cresciuta in coltura aeroponica

Il presente contributo riporta i risultati riguardanti la biofortificazione dell’avena col fine ultimo di aumentare il contenuto totale di ferro nella pianta. Questa pianta, come molte altri monocotiledoni, assorbe il ferro tramite dei composti essudati dalle radici e conosciuti come fitosiderofori. Al fine di aumentare l’assorbimento di ferro, le piante sono lasciate crescere in soluzione nutritiva priva di ferro e quest’ultimo è aggiunto in un secondo momento sotto forma in ferro inorganico (ferro (III) ammonio solfato). Questo trattamento è quindi confrontato con la crescita standard con ferro chelato (Fe-EDDHA). Sono anche stati investigati tempi diversi di crescita senza ferro e diversi tempi di raccolta.

**Key words**: Biofortification, iron, oats, phytosiderophores, aeroponic growth.

# **1. Introduction**

The aim of this part of the PhD project is to enhance the nutritional content of an already established microgreen and in particular its iron content. Oat (*Avena sativa* cv. “Prevision”) was chosen because its seeds are a well-known source of iron and is also gluten-free, making it suitable as integrator for many kinds of diets. The results of experiments involving iron starvation to force the release of phytosiderophores indicate that this strategy can be successful to biofortify oats. Economical and production advantages are also briefly discussed in view of the shortening of growing/harvesting cycles that was explored in a second series of experiments. Additionally, it is hypothesized that this growing approach can be used for biofortification of other minerals or in co-culturing systems to increase iron content in other iron-inefficient plants.

# **2. Materials and methods**

Seeds of oats (*Avena sativa* cultivar “Prevision”) were bought by a commercial seed producer. Plants were grown either at the aeroponic plant of Zero s.r.l. in Pordenone or hydroponically at the lab in Conegliano. “Starvation” experiments were conducted with plants grown for different amounts of time either on a modified Hoagland nutritive solution without iron, the same nutritive solution with iron chelate or simple osmotized water. When inorganic iron was added, it was in the form of iron (III) ammonium sulfate whereas the iron chelate was Fe-EDDHA. Hydroponic growth was performed under comparable parameters, but the nutritive solution was constantly aerated using an aquarium pump and aeration stones.

Plant material was desiccated using a desiccator oven for the time needed to reach constant weight (usually 3-4 days). The desiccated plants were ground to a fine powder using a coffee grinder and mineralized using a combination of sulfuric acid and hydrogen peroxide. The obtained mineralized samples were analysed by AAS.

# **3. Results and discussion**

**Table 2**: *Avena sativa* cv. “Prevision” iron content under different growing conditions. Iron was supplied as inorganic iron at 11 mg/L

|  |  |
| --- | --- |
| Treatment type | Iron content(mg/100g DW) |
| 7 days iron-free nutritive solution |  |
|  1 day after iron addition | 15,9 |
|  2 days after iron addition | 23,8 |
|  4 days after iron addition | 37,2 |
| 7 days osmotized water |  |
|  1 day after iron addition | 38,4 |
|  2 days after iron addition | 43,0 |
|  4 days after iron addition | 60,9 |

**Table 1**: *Avena sativa* “Scura” iron content under different iron fertilization

|  |  |
| --- | --- |
| Treatment type | Iron content(mg/100g DW) |
| 7 days no iron | 10,6 |
|  + 5 days 1,2 mg/L Fe-HBED | 13,6 |
| 7 days 1,2 mg/L Fe-HBED | 11,3 |
|  + 5 days 1,2 mg/L Fe-HBED | 16,7 |
| 8 days 1,2 mg/L Fe-DTPA | 14,2 |

Exploratory experiments have demonstrated that the addition of iron in the form of iron chelate (Fe-HBED and Fe-DTPA) has little to no effect on the amount of iron found in the plant (Table 1). These results are in accordance with the literature (Jolley and Brown, 1989b), which shows that oat uptake of iron in the form of chelates is less efficient. Similarly, it was noted that when plants were grown under iron starvation they would show a high uptake capacity from inorganic sources of iron (Reid et al., 1989).

Following the experimental setup of Reid and colleagues, oats were grown for 7 days in osmotized water or nutritive solution without iron. After this time iron was added as inorganic iron. Plants starved in this manner showed a marked increase in iron content (Table 2).

In another set of experiments, plants were grown in standard nutritive solution (containing Fe-EDDHA), iron-free nutritive solution or osmotized water. A longer timeframe was allowed to pass before iron addition and post-addition harvesting was also delayed longer. Also in this case plants starved for iron showed a much higher iron content. Additionally the experiment confirmed that iron nutrition through chelates is ineffective (Table 3).

These results set a promising framework for the efficient and minimally invasive biofortification of oat microgreens. Varying the timeframe has proven that shorter cycles are effective as much as longer ones. Shortening the growing and harvesting periods leads to lower expenditure for lighting, lower risk of contamination during growth and higher number of growing cycles per year. Furthermore, increasing the amount of inorganic iron to 22 mg/L didn’t cause any appreciable difference from the fertilization at 11 mg/L, allowing a reduction of inorganic fertilizer needed.

**Table 3**: *Avena sativa* cv. “Prevision” iron content under different growing conditions. Iron was supplied as Fe-EDDHA at 1,4 mg/Lor as inorganic iron at 22 mg/L

|  |  |
| --- | --- |
| Treatment type | Iron content(mg/100g DW) |
| 10 days standard nutritive solution |  |
|  4 days after | 6,9 |
|  10 days after | 7,3 |
| 10 days iron-free nutritive solution |  |
|  4 days after iron addition | 16,0 |
|  10 days after iron addition | 21,1 |
| 10 days osmotized water |  |
|  4 days after iron addition | 64,7 |
|  10 days after iron addition | 62,4 |

Since phytosiderophores have varying affinities for several other elements (calcium, zinc, magnesium etc.) it is of further interest to investigate if the “starvation” setup can be utilized to force the uptake of other minerals of interest. Another path for biofortification is the co-culturing of crop plants, which has been already tested in hydroponic systems (Cesco et al., 2006). In this case oat plants are grown together with other iron-inefficient plants to enhance the iron uptake of the latter plants. Alternatively, a phytosiderophore-rich nutritive solution can be used as a base for growing iron-inefficient plants, constituting a form of chelated iron fertilizer that is totally of plant origin.

# **4. References**

Cesco S., Adamo D.R, Tagliavini M., Varanini Z, Pinton R. (2006) Phytosiderophores released by graminaceous species promote 59Fe-uptake in citrus. *Plant Soil* **287**:223–233

Jolley V.D. and Brown J.C. (1989a) Iron efficient and inefficient oats. I. Differences in phytosiderophore release. *Journal of Plant Nutrition*, **12**:423-435

Jolley V.D. and Brown J.C. (1989b) Iron inefficient and efficient oat cultivars. II. Characterization of phytosiderophore released in response to Fe deficiency stress. *Journal of Plant Nutrition*, **12**:923-937

Reid C.P.P., Crowley E., Kim J., Powell P.E. and Szaniszlo P.J. (1984) Utilization of iron by oat when supplied as ferrated synthetic chelate or as ferrated hydroxamate siderophore. *Journal of Plant Nutrition*, **7**:437-447