Characterization of iron-deficiency responses and targeted knockout of the pgrl1 gene in *Physcomitrella patens*

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In *Arabidopsis thaliana* and *Chlamydomonas reinhardtii* iron-deficiency leads to chlorosis, remodeling of photosynthetic apparatus and induction of the iron uptake systems (*irt1, fro, fre*) [2] as well as to induction of the thylakoid transmembrane protein PGRL1 [1, 3]. Phylogenetic genomic profiling sorted the *pgrl1* gene to a class of genes called “plastid cut”, which contains no more than 90 genes that are highly conserved from diatoms to vascular plants [4]. In *Arabidopsis thaliana* PGRL1 appears to be involved in cyclic photosynthetic electron transfer (CEF) [5]. In *Chlamydomonas reinhardtii* PGRL1 is required for efficient CEF under iron-deprivation [6].

In the present study, the iron-deficiency responses and the function of PGRL1 were analysed in the moss *Physcomitrella patens*. Loss of chlorophyll binding proteins, a decrease in the chlorophyll a/b ratio and decreased abundance of Photosystem I proteins were observed in iron-deficient protonema after 12-16 days growth under iron-deficient conditions. Furthermore, qRT-PCR revealed, decreased level of *fer* (ferritin) mRNA, increased level of the transcript of the ferric chelate reductase oxidase (*fro*) and unchanged expression of the *irt1* gene (encoding the divalent iron transporter). Moreover the mRNA expression level of *pgr5* and *pgrl1* were induced in response to iron-deficiency, suggesting an important function of both genes in iron-dependent processes in *P. patens.*

To study the function of PGRL1 in *P. patens, pgrl1*-knockout cassette was generated and transformed using two methods: particle bombardment and PEG mediated protoplasts transformation. Stable mutants as well as mutants that expressed the transgene transiently or extrachromosomally were obtained. Currently the mutant population is screened for transformants that harbor a *pgrl1* gene knock-out.

References:
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