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Signals from chloroplasts and mitochondria for iron homeostasis regulation

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Iron (Fe) is an essential element for human nutrition. Given that plants represent a major dietary source of Fe worldwide, it is crucial to understand plant Fe homeostasis fully. A major breakthrough in the understanding of Fe sensing and signaling was the identification of several transcription factor cascades regulating Fe homeostasis. However, the mechanisms of activation of these cascades still remain to be elucidated. In this opinion, we focus on the possible roles of mitochondria and chloroplasts as cellular Fe sensing and signaling sites, offering a new perspective on the integrated regulation of Fe homeostasis and its interplay with cellular metabolism.

Highlights

► The understanding of plant iron homeostasis must be fully elucidated. ► We review the knowledge of metabolic adaptation in response to altered iron availability. ► Mitochondria and chloroplasts are proposed to act as cellular iron sensing and signaling sites.

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Metabolic adaptation to the Fe status of a plant

Fe is essential for most living organisms, and plants represent the main dietary source of Fe in human nutrition. Consequently, Fe uptake by roots, its distribution throughout organs, tissues and subcellular compartments (Boxes 1 and 2), the synthesis of prosthetic groups, such as heme or Fe–sulfur (S) clusters, and Fe storage were major focuses of scientific research during the past decade (for overview see [1]). Fe is essential for vital metabolic reactions in organelles, such as respiratory and photosynthetic electron transport chains (RET and PET, respectively); thus, its imbalance affects the entire cellular metabolism. To optimize effective Fe acquisition, distribution, and utilization, plants react to changes in Fe availability in their environment by adapting their metabolism and by tightly regulating Fe homeostasis. Despite the wealth of knowledge that has been gained concerning the processes by which plants can respond to Fe deficiency, the mechanisms of Fe sensing and signaling are not yet fully understood. So far, several key players in the major transcriptional networks that control Fe homeostasis have been identified, as well as several different Fe-responsive signaling pathways [2] (Box 1).

Box 1

Plant Fe-transport mechanisms and transcriptional control

In plants, the main obstacle to overcome concerning Fe nutrition is related to its low availability in the soil. In fact, notwithstanding its abundance, Fe exists in well-aerated soils mainly as scarcely soluble Fe(III)-oxides and oxihydroxides and therefore is not freely available for uptake by plants. To overcome with this situation, plants have evolved efficient mechanisms to acquire Fe from the soil. Dicots and nongrass plants developed the so-called 'Strategy I' mechanism, by which Fe acquisition is mediated via a reduction-based mechanism [2]: a ferric chelate reductase converts the Fe in Fe(III)-chelates to free Fe²⁺ and, subsequently, an Fe²⁺ transporter (IRT1, IRON-REGULATED TRANSPORTER 1) moves Fe across the plasma membrane into the cell. Additionally, in most of the Strategy I plants studied so far, there is an associated increase in the activity of a plasma membrane H⁺-ATPase, which actively extrudes protons. Extracellular protons in turn are necessary both for decreasing rhizospheric pH to solubilize Fe(III) and for generating the electrochemical proton gradient to drive Fe uptake.

By contrast, in grasses (Strategy II plants), a group that includes most of the staple grain crops worldwide, a distinct mechanism for Fe uptake has evolved. These plants produce molecules of the mugineic acid family called phytosiderophores (PSs). PSs are secreted into the rhizosphere by the TOM1 (transporter of mugineic acid family phytosiderophores 1) transporter where they chelate and help to solubilize Fe³⁺. The Fe(III)–PS complex is then taken up into root cells through the action of YELLOW STRIPE 1 (YS1) proteins [2,15,77].

To sustain Fe uptake, plants reprogram the entire metabolism. In particular, in Strategy I plants, Fe deficiency strongly affects mitochondrial functionality and, therefore, cells induce alternative energetic pathways, such as glycolysis, oxidative pentose phosphate pathways, or fermentation [36]. In addition, in Strategy II plants, Fe deficiency increases some substitute energetic pathways, although no specific data are available concerning the effect on mitochondria [11]. To increase Fe availability and acquisition, as well as production of mugineic acids (Strategy II), synthesis of phenols (Strategy I) and of organic acids (both Fe-uptake mechanisms) is induced.

Progress has been made in identifying and characterizing several transcription factors (TFs) that regulate plant Fe-deficiency responses and which have been reviewed recently [2,15,78]. These TFs act by trans-activating primary target genes, such as Fe(III)-chelate reductases, biosynthetic enzymes of the mugineic acid Fe-chelators, and transporters. Briefly, in dicotyledonous plants, the regulation of Strategy I genes is under the control of two main and independent transcriptional pathways: the FER-LIKE IRON-DEFICIENCY-INDUCED TRANSCRIPTION FACTOR (FIT) and the POPEYE-mediated networks. The FIT-mediated network is mainly required for the regulation of the Fe-deficiency response at the epidermal cell level, whereas the POPEYE-mediated network occurs in the vasculature and regulates genes implicated in metal homeostasis. In grasses, the regulation of Strategy II genes requires the TFs OsIDEF1 (IDEF, iron deficiency-responsive element-binding factor) and OsIDEF2, as well as OsIRO2, as positive regulators of the Fe-deficiency response in rice.

Box 2

Organellar Fe transport

Once Fe enters the plant cell, it may be either integrated in cytoplasmic proteins or diverted to chloroplasts, mitochondria, or vacuoles. PERMEASE IN CHLOROPLASTS 1 (PIC1) is involved in Fe transport to chloroplasts [79,80], whereas the MITOCHONDRIAL IRON TRANSPORTER (MIT) loads cytoplasmic Fe to mitochondria [19,81]. Vacuoles, serving as additional Fe pools, acquire Fe through the VACUOLAR IRON TRANSPORTER 1 (VIT1; [82]). For chloroplasts, nuclear genes of primary photosynthesis are reciprocally regulated in Arabidopsis knockout and overexpressing mutants of the Fe-uptake protein PIC1 [79,80]. Interestingly, although photosynthesis-associated transcripts accumulate in PIC1-overexpressing lines, plants suffer from chlorosis and reduced growth, most likely due to plastid-internal Fe toxicity. Thus, PIC1ox plants show an antagonist effect of photosynthetic activity and regulation of gene expression, similar to the mentioned reciprocal Fe-deficiency regulation of mitochondrial RET activity and transcript and/or protein abundance (see main text). It is therefore tempting, but still preliminary, to speculate whether organellar retrograde signals, which might be generated by metabolic and/or activity changes in key proteins, might compete with cytosolic Fe sensing, which would trigger an anterograde pathway (Box 3).

Plants could sense Fe deficiency and/or toxicity in the cytosol, and excess Fe could either be diverted into the vacuole or into plastids, where it could be stored in the Fe-storage protein ferritin. An indication for cytosolic and nonplastid Fe sensing is provided by knockout mutants of PIC1, which show a strong increase in ferritin transcripts and proteins [79], possibly reflecting a rise in cytosolic Fe, while Fe uptake into chloroplasts is blocked. A reduction in mitochondrial Fe uptake in rice is suggested to induce Fe excess in the cytoplasm as well as the upregulation of OsVIT1[19], additionally supporting cytosolic Fe perception. However, the mechanism of this sensing is not clearly understood. The importance of transporter-mediated Fe trafficking between the various subcellular compartments has been further documented in Arabidopsis seeds. Manipulation of Fe levels by overexpression of AtVit1 or knocking out Nramp3 and Nramp4 (two transporters responsible for Fe unloading from the vacuole) result in a decrease in ferritin abundance in the plastids [83], in this case arguing for cytosolic Fe sensing and thus anterograde signals.

In short, cellular transporters are regulated in response to Fe deficiency and/or excess and work in close coordination to manage changes in Fe availability, and to tune the appropriate delivery of the metal. Therefore, they represent important targets and tools for future studies on the signaling of plant Fe-homeostasis regulation.

In response to Fe deficiency, plants initiate several metabolic changes to bring about increased Fe acquisition capacity, as extensively reviewed previously [3–5]. On the one hand, a low Fe content leads to a high energy request to sustain the increased rate of Fe acquisition; on the other hand, it limits the functioning of mitochondria and chloroplasts, which are Fe-dependent energy-producing organelles. To address this energetic emergency, cells must increase the rate of energy-producing alternative pathways. These increases have been observed in both Strategy I [6–10] and Strategy II plants [11] (Box 1). Because the photosynthetic apparatus is strongly affected under Fe deficiency, the existence of a carbon flow from root to leaf in the form of carboxylates has been observed in Fe-deficient plants [12,13].

The metabolic reprogramming under Fe deficiency relates to the functional alteration of essential cell compartments, such as mitochondria and chloroplasts. Organellar processes could therefore be involved in Fe sensing. Among them, Fe distribution among the various subcellular compartments is crucial and is controlled by organellar Fe transporters [14–16], as described in Box 2.

Fe deficiency-induced changes in plant mitochondria and chloroplasts

Mitochondria are a major subcellular compartment for Fe metabolism, because Fe is an essential cofactor for several proteins belonging to RET and the tricarboxylic acid (TCA) cycle. Moreover, part of Fe–S cluster and heme biosynthesis is located in the mitochondria and is strictly linked to Fe homeostasis. Hence, alteration of the cellular Fe status dramatically influences mitochondrial functionality and thereby cellular metabolism [3,4,17–19].

Fe deficiency decreases RET activity [20]. However, Fe-deficient *Arabidopsis* (*Arabidopsis thaliana*) roots show accumulation of some respiratory transcripts and proteins [21,22], suggesting the existence of antagonistic levels of regulation under Fe starvation. Furthermore, several authors have reported that, under Fe deficiency, induction of TCA cycle activity leads to the strong production of organic acids, which are intermediate compounds of the TCA cycle [4]. Because succinate dehydrogenase and aconitase are Fe–S proteins, it has recently been hypothesized that, under Fe deficiency, the TCA cycle might shift to a linear mode, bypassing the affected enzymes [4]. Furthermore, it appears that aconitase is affected by metal-induced damage [23] and that inhibition of aconitase activity by reactive oxygen species (ROS, see below) leads to mitochondrial citrate export [24].

Similarly to mitochondria, chloroplasts also have several Fe-containing proteins located in all plastid subcompartments, with the most prominent functioning in PET [5]. Fe shortage thus induces a decrease in the pigment content (chlorophyll and carotenoids) and in proteins belonging to the photosynthetic apparatus. As a result, the photosynthetic rate diminishes, as well as the amount of the photosystem II (PSII) antennae, which exhibit an altered aggregation, concomitant with a decrease in xanthophyll content [25–29]. Accordingly, in the green algae *Chlamydomonas* (*Chlamydomonas reinhardtii*) a decrease in the amount of photosystem I (PSI), as well as a remodeling of the PSI-associated light-harvesting complex I (LHCI), were observed [30,31]. Other photosynthesis-related proteins also decrease in abundance, such as components of inorganic carbon assimilation, plastocyanin, proteins associated with the light reaction, and enzymes belonging to the Calvin cycle [32], suggesting that Fe deficiency affects the protein abundance of the photosynthetic apparatus both at the thylakoid as well as the stromal and luminal levels [30,33].

Mitochondria and chloroplasts in Fe-dependent oxidative stress signal production and nitric oxide-mediated control of Fe homeostasis

Iron imbalance is known to induce oxidative stress in cells by promoting the generation and accumulation of ROS, causing oxidation of cellular components, hindering metabolic activities, and affecting organelle integrity [34]. Indeed, Fe is a cofactor of many antioxidant enzymes, including catalase, peroxidases, ascorbate peroxidase, and Fe-superoxide dismutase. These enzymes are strongly affected by Fe deficiency, leading to a consistent unbalance of the detoxification processes in plants, which thus accumulate ROS [35,36]. Fe is also a pro-oxidant factor that can catalyze free radical production in the presence of reductants and peroxides. In particular, Fe-mediated ROS production is involved in the transcriptional regulation of the expression of ferritin genes, encoding chloroplast proteins storing Fe when in excess [37].

PET and RET activities determine the redox states of their respective organelles, being associated with reducing power or energy-carrying metabolites, such as NAD(P)H and ATP [38–40]. The organellar redox state can be monitored by the production of ROS, which in turn is associated with changes in nuclear gene expression (NGE) [41]. The role of ROS as retrograde signals has been questioned, because they are probably too short lived to reach the nucleus and too unspecific to act as information carriers. Nevertheless, it has been hypothesized that ROS might act indirectly by generating specific peptides from the proteolytic breakdown of oxidized proteins that might have signal function, contributing to retrograde signaling and gene regulation, as observed in mitochondria [42]. Furthermore, under Fe deficiency, the altered RET and PET could lead to ROS overproduction and, thus, to protein oxidation (Figure 1) [43–45]. Alteration of the assembly of the protein complexes of RET and PET under Fe deficiency (see above) could produce peptides hosting Fe–S clusters, which might be oxidized and/or degraded and then function as signals.

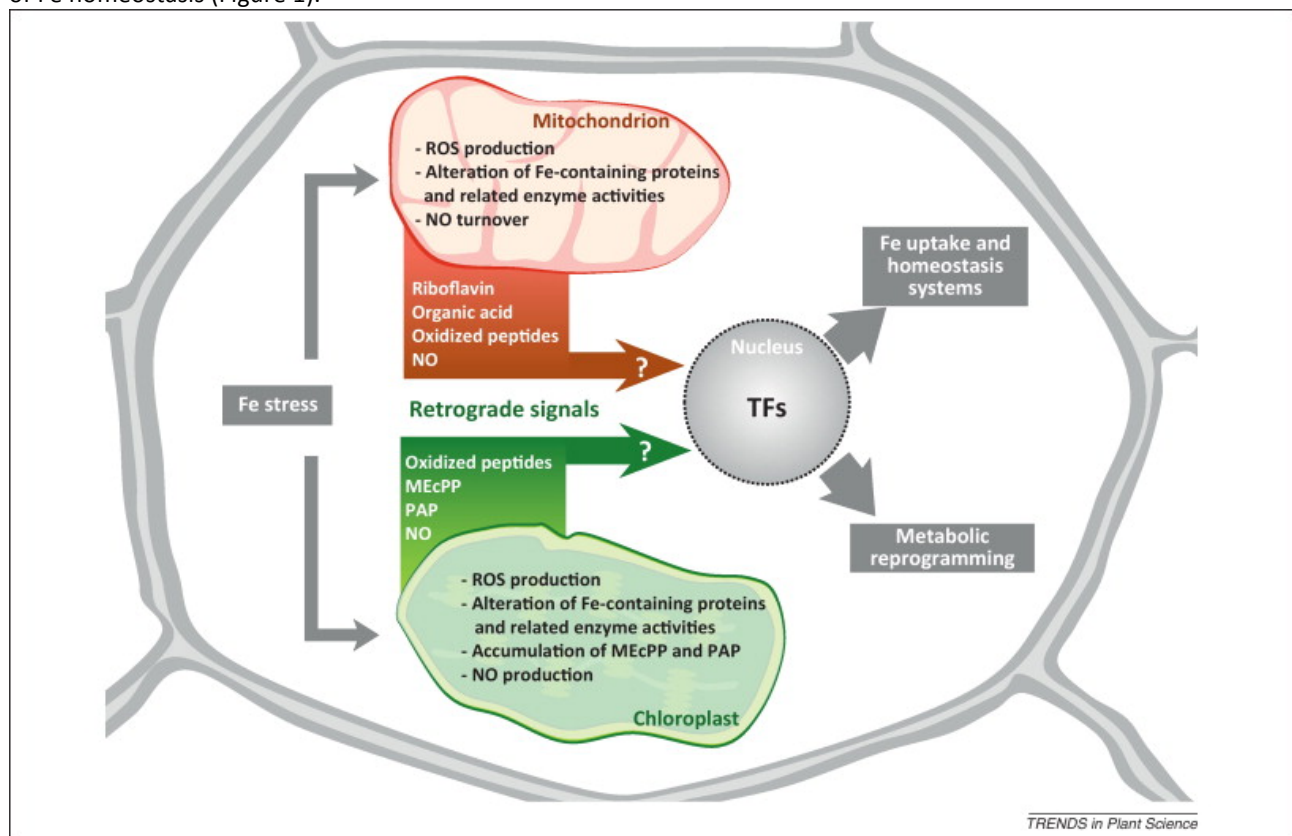
Hypothetical retrograde signals produced by mitochondria and chloroplasts under...

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Figure 1. Hypothetical retrograde signals produced by mitochondria and chloroplasts under iron (Fe) stress conditions and their possible implication in Fe sensing and signaling systems in plants. Fe-stress conditions affect both respiratory (RET) and photosynthetic (PET) electron transport chains in mitochondria and chloroplasts. Furthermore, these conditions lead to the production of reactive oxygen species (ROS), which in turn might oxidize peptides. These

oxidized peptides could function as specific retrograde signals. As described in the main text, in both organelles, Fe-containing proteins and enzymes can be affected by Fe stress. Furthermore, an accumulation of methylerythritol cyclodiphosphate (MEcPP) and 3'-phosphoadenosine 5'-phosphate (PAP) in chloroplasts, and export of riboflavins and organic acids in mitochondria might occur in response to Fe stress. Nitric oxide (NO) represents another important signal molecule involved in the adaptation of Fe homeostasis. In chloroplasts, Fe excess promotes NO production, whereas in mitochondria, NO turnover is most likely to be involved in regulation. Furthermore, mitochondria and chloroplast metabolism might act directly as cellular Fe sensors via modification of enzymes. Subsequently, they could produce retrograde signals responsible for the recruitment and/or activation of the transcription factor (TF) cascades responsible for the regulation of Fe uptake and homeostasis systems and for tuning of cellular metabolism.

An important signal molecule involved in the establishment of Fe homeostasis in plants is nitric oxide (NO), which acts downstream of the Fe transcription factor FIT [46–50]. Cellular NO production involves both mitochondria and chloroplasts. Plant mitochondria participate in NO turnover by the interconversion of nitrite and NO at the site of complex IV [51], and plastids are important players in regulating NO levels in plant cells [52]. Furthermore, Fe excess induces NO production within the chloroplasts before the upregulation of expression of the ferritin AtFer1 gene [46,53], thereby reinforcing the idea that retrograde signaling by organelles could play a crucial role in the regulation of Fe homeostasis (Figure 1).



Putative retrograde signals from Fe-stressed plastids and mitochondria

The coordination between organelle gene expression (OGE) and NGE requires both anterograde and retrograde signals [54] (Box 3). The retrograde pathway communicates changes in the metabolic status of the organelles to the nucleus and consequently changes the anterograde signals. Changes in the metabolic status of chloroplasts and/or mitochondria (e.g., in response to an altered Fe nutritional status) have profound effects on the rest of the plant cell, and involve massive changes in the transcript profiles of nuclear genes. Despite the evidence that the main cellular targets under Fe deficiency are mitochondria and chloroplasts, no specific study has yet investigated the role of these organelles in Fe sensing and signaling in plants. It is therefore timely to investigate hypothetical retrograde signals from chloroplasts and mitochondria as a result of Fe stress.

Box 3

Anterograde and retrograde signaling pathways

In response to endogenous and environmental stimuli that are perceived by the nucleus, an anterograde mechanism transmits signals originating from the nucleus to coordinate gene expression in organelles. The regulation of OGE mostly occurs through post-transcriptional mechanism and involves nuclear-encoded proteins, acting as regulators. By contrast, retrograde mechanisms transmit signals originating in the organelles to regulate NGE, which can then alter anterograde control. These mechanisms allow the communication of the functional and developmental state of organelles to the nucleus, which can thus modulate cellular metabolism [84]. In photosynthetic organisms, this regulation is more complex owing to crosstalk between mitochondria and chloroplasts. Tight coordination between the nucleus and organelles is crucial to the survival of eukaryotic cells: not only are chloroplasts and mitochondria of great bioenergetic importance, but they also synthesize many different cellular metabolites, including amino acids, fatty acids, isoprenoids, nucleotides, vitamins, and porphyrins.

For example a 'biogenic' retrograde signal is used by plastids as they develop into chloroplasts in young photosynthetic tissue. When chloroplast development is blocked, the expression of nuclear genes involved in photosynthesis and chloroplast biogenesis is greatly reduced. Arabidopsis genomes uncoupled (*gun*) mutants still express these nuclear genes even when chloroplast development is impaired ([85] and references therein). *gun* mutants demonstrate the implication of the branch point of the heme–chlorophyll biosynthesis pathway in the plastid as source of retrograde signals. The protoporphyrin IX precursor for heme is known to exit the plastid and is a mitochondrial retrograde signal in yeast. However, the mechanisms of its transport to the cytoplasm and of its interaction with signaling pathways are unknown. 'Operational' retrograde signals are also produced by plastids in response to external cues and stress. In contrast to *gun* signals, operational signals control nuclear genes to limit and repair damage from ROS generated by abiotic stresses, such as excess light and drought [54,85].

Plastid retrograde signals could derive from various sources, including the tetrapyrrole pathway, the level of ROS, or the redox state of the plastids [55]. All these signals are thought to converge on the regulator GENOMES UNCOUPLED 1 (*GUN1*) [56]. Recently, a new circadian Fe-sensing system located within chloroplasts has been identified in Arabidopsis [57,58]. It is independent of *GUN1*, requires an uncharacterized plastid-encoded protein, and depends on both the functional state of the chloroplast and the phytochromes [57]. On the one hand, these findings suggest that the known retrograde signals do not directly participate in Fe sensing mechanisms in plants. On the other hand, they emphasize the role of plastids in Fe sensing and signaling in a retrograde way. This raises the need to identify retrograde signal molecules potentially involved in plastid-to-nucleus signaling for tuning Fe homeostasis.

In this context, new plastid retrograde signals have been recently identified, such as the metabolites methylerythritol cyclodiphosphate (MEcPP) [59], and 3'-phosphoadenosine 5'-phosphate (PAP) [60]. MEcPP is an intermediate of the plastid methylerythritol phosphate (MEP) pathway, suggested to act as an NGE regulator [59]. 1-Hydroxy-2-methyl-2-(E)-butenyl-4-diphosphate synthase (HDS) is a bottleneck enzyme in this pathway. In an Arabidopsis mutant defective in the *CEH1* gene encoding HDS, MEcPP accumulates, the expression of selected stress-responsive genes increases, and salicylic acid accumulation occurs. Abiotic stresses, such as high light or wounding, also elevate MEcPP levels. It was proposed that the increased level of MEcPP serves as a specific and critical retrograde signal eliciting the expression of selected stress-responsive nuclear-encoded and plastid-localized proteins. Because HDS is a Fe–S cluster protein [61], it can be postulated that Fe-deficiency conditions could lead to MEcPP accumulation and, thus, the MEP pathway could be a metabolic stress sensor during Fe deficiency (Figure 1).

PAP is known to accumulate in Arabidopsis plastids in response to high light or drought stress and was able to regulate stress-responsive nuclear genes [60]. It accumulated in a *sal1* mutant deficient in the enzyme able to dephosphorylate PAP to AMP. The mode of action of PAP could be inhibition of 5' to 3' exonucleases. Transcriptome analysis of Arabidopsis *sal1* and *xrn4* mutant plants (supplemental material in [60]) further revealed that the ferritin genes *AtFer1*, *AtFfer3*, and *AtFer4* are upregulated in these genetic backgrounds, thus establishing a link between the PAP retrograde signaling pathway and the regulation of Fe homeostasis genes (Figure 1).

Mitochondrial retrograde regulation in plants is poorly understood and few signaling pathways have been identified [62]. As stated above, mitochondria are a crucial cellular site for the synthesis of Fe–S clusters, both for themselves and for Fe–S proteins from the cytosol and the nucleus [63]. Thus, impaired Fe–S cluster assembly in the mitochondria could influence or could be used as a sensor to signal alterations in Fe homeostasis. Some evidence supporting this hypothesis is found in yeast. In *Saccharomyces cerevisiae*, the expression of Fe uptake and storage genes is controlled by the Fe-responsive transcription factors *Aft1* and *Aft2* [64,65], which accumulate in the nucleus under Fe deficiency

and activate the Fe regulon. The activation of Aft1/2 is mediated by the so-called 'Fra-Grx iron signaling pathway', which includes: (i) the mitochondrial Fe-S cluster biosynthesis machinery; (ii) the glutaredoxines Grx3 and Grx4; and (iii) an aminopeptide P-like protein named Fe repressor of activation-1 (Fra1) [66,67]. Under Fe-replete conditions, the Fra-Grx signaling pathway inhibits the Aft1/2 activity. By contrast, under Fe-deprived conditions, the affected mitochondrial Fe-S cluster biogenesis leads to the deactivation of the Fra-Grx pathway, allowing the Atf1 transcription factor to activate the Fe regulon [68]. Furthermore, frataxin, a mitochondrial protein involved in Fe homeostasis, might act as a mitochondrial sensor of the Fe content, regulating the rate of Fe-S biogenesis [69,70]. These findings suggest that molecules deriving from mitochondria, such as Fe-S clusters, are components of a retrograde pathway operating under Fe deficiency. Moreover, it has been suggested that some Fe-dependent metabolites (such as those provided by Fe-dependent enzymes) and/or heme contents are key regulators of central transcription factors, inducing the Fe regulon in yeast [71]. Furthermore, in Fe-deficient plants, accumulation of riboflavins has been observed [9,72]. These compounds are cofactors of complexes I and II of RET. It was therefore postulated that, by affecting RET-complexes, Fe deficiency allows the unused riboflavins to be transported outside the mitochondria [4,73]. Considering that riboflavins are also Fe³⁺-chelate reductase cofactors and that their accumulation is controlled by transcription factors regulating Fe assimilation [74], they might be considered as signal molecules of the physiological mitochondrial status under low Fe.

Concluding remarks and outlook

Despite great progress in understanding Fe homeostasis in plants, little insight has been gained concerning the effects of Fe nutritional status on the metabolism of organelles, and the consequences for Fe signaling.

Both Fe-deficient or Fe-excess conditions are known to affect mitochondrial and chloroplast metabolism through post-transcriptional and/or translational modifications of key enzymes. Such modifications are fast and enable a quicker response to environmental changes than via transcriptional activation. In particular, in the case of Fe deficiency, activity of Fe-containing enzymes is negatively regulated and, consequently, corresponding metabolite pools could be modified. Thus, we hypothesize that organelle retrograde signals could be produced from these post-transcriptional and/or post-translational-mediated metabolic changes, and transduced for subsequent regulation of nuclear genes that are important for Fe uptake and homeostasis, via the activation of the already established cascade of transcription factors (Figure 1). According to this scenario, long-term cellular metabolic reprogramming would accompany and follow adaptation of Fe uptake and homeostasis.

A challenge is to answer the question of whether Fe-mediated modifications of mitochondrial and chloroplast metabolism in response to alteration of the Fe status represent a source of retrograde signals, necessary to regulate the NGE involved in Fe stress responses. Therefore, there is a need to document kinetically the metabolic changes occurring in mitochondria and chloroplasts in response to Fe deficiency or excess, and the crosstalk between these organelles during these changes. Furthermore, it would also be interesting to study the organellar crosstalk in root cells, where nonphotosynthetic plastids are present, and compare it with crosstalk in leaf cells. Indeed, in root cells, where the plant manages the acquisition of Fe, plastid-mitochondrial crosstalk would generate a root-specific signal pool able to regulate NGE of the roots. In addition, in leaf cells, where the plant manages the synthesis of organic compounds, chloroplast-mitochondria crosstalk would generate a leaf-specific signal pool. Whether the integration of these root and leaf signal pools contributes to the long-distance signaling of Fe deficiency [75] is an attractive question for future research. Whether sugars, or other molecules of general metabolism, which are transported in the phloem, participate in this systemic signal [75], is still unknown. However, in roots of some Fe-deficient plants, a change in the carbohydrate metabolism was observed with an increase in the raffinose family of oligosaccharides (RFOs) [9]. It has been reported that oligosaccharides, mainly generated from cell wall damage, might act as signaling molecules under some stress conditions [76] and that the RFOs might act as long-distance Fe-deficiency signals via phloem sap transport [9].

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References

1

J. Balk, M. Pilon

Ancient and essential: the assembly of Fe-S cluster in plants

Trends Plant Sci., 16 (2011), pp. 218-226

ArticlePDF (533KB)View Record in Scopus

2

M.N. Hindt, M.L. Guerinot

Getting a sense for signals: regulation of the plant iron deficiency response

Biochim. Biophys. Acta, 1823 (2012), pp. 1521-1530

ArticlePDF (339KB)View Record in Scopus

3

G. Zocchi

MetaG. Vigani, G. Zocchi

The fate and the role of mitochondria in Fe-deficient roots of Strategy I plants

Plant Signal. Behav., 2009 (2009), pp. 375-379

CrossRefView Record in Scopus

9

R. Rellán-Álvarez, et al.

Changes in the proteomic and metabolic profiles of Beta vulgaris root tips in response to iron deficiency and resupply

BMC Plant Biol., 10 (2010), p. 120

CrossRef

10

S. Donnini, et al.

Proteomic characterization of iron deficiency responses in Cucumis sativus L. roots

BMC Plant Biol., 10 (2010), p. 268

CrossRef

11

A.F. López-Millán, et al.

Carboxylate metabolism change induced by Fe deficiency in barley, a Strategy II plant species

J. Plant Physiol., 169 (2012), pp. 1121-1124

ArticlePDF (209KB)View Record in Scopus

12

J. Abadía, et al.

Organic acids and Fe deficiency: a review

Plant Soil, 241 (2002), pp. 75-86

CrossRefView Record in Scopus

13

J. Abadía, et al.

Towards a knowledge-based correction of iron chlorosis

Plant Physiol. Biochem., 49 (2011), pp. 471-482

ArticlePDF (1MB)View Record in Scopus

14

K. Bashir, et al.

Iron uptake and loading into rice grains

Rice, 3 (2010), pp. 122-130

CrossRefView Record in Scopus

15

T. Kobayashi, N. Nishizawa

Iron uptake, translocation and regulation in higher plants

Annu. Rev. Plant Biol., 63 (2012), pp. 131-152

CrossRefView Record in Scopus

16

S.S. Conte, E.L. Walker

Transporters contributing to iron trafficking in plants

Mol. Plant, 4 (2011), pp. 464-476

ArticlePDF (343KB)CrossRefView Record in Scopus

17

M.V. Busi, et al.

Deficiency of Arabidopsis thaliana frataxin alters activity of mitochondrial Fe-S proteins and induces oxidative stress

Plant J., 48 (2006), pp. 873-882

CrossRefView Record in Scopus

18

M.V. Maliandi, et al.

The mitochondrial protein frataxin is essential for heme biosynthesis in plants

FEBS J., 278 (2011), pp. 470-481

CrossRefView Record in Scopus

19

K. Bashir, et al.

The rice mitochondrial iron transporter is essential for plant growth

Nat. Commun., 2 (2011), p. 322

CrossRef

20

G. Vigani, et al.

Iron availability affects the function of mitochondria in cucumber roots

New Phytol., 182 (2009), pp. 127-136

CrossRefView Record in Scopus

21

O. Thimm, et al.

Response of Arabidopsis to iron deficiency stress as revealed by microarray analysis

Plant Physiol., 127 (2001), pp. 1030-1043

CrossRefView Record in Scopus

22

P. Lan, et al.

iTRAQ protein profile analysis of Arabidopsis roots reveals new aspects critical for iron homeostasis

Plant Physiol., 155 (2011), pp. 821-834

CrossRefView Record in Scopus

23

Y.F. Tan, et al.

Divalent metal ions in plant mitochondria and their role in interactions with proteins and oxidative stress-induced damage to respiratory function

Plant Physiol., 152 (2010), pp. 747-761

CrossRefView Record in Scopus

24

M.J. Morgan, et al.

Decrease in manganese superoxide dismutase leads to reduced root growth and affects tricarboxylic acid cycle flux and mitochondrial redox homeostasis

Plant Physiol., 147 (2008), pp. 101-114

CrossRefView Record in Scopus

25

H. Marschner (Ed.), Mineral Nutrition of Higher Plants, Academic Press (1995)

26

S. Merchant, et al. (Eds.), The Structure and Function of Plastids, Springer (2007)

27

A.M. Timperio, et al.

Proteomics, pigment composition, and organization of thylakoid membranes in iron-deficient spinach leaves

J. Exp. Bot., 58 (2007), pp. 3695-3710

CrossRefView Record in Scopus

28

S. Donnini, et al.

Differential responses in pear and quince genotypes induced by Fe deficiency and bicarbonate

J. Plant Physiol., 166 (2009), pp. 1181-1193

[ArticlePDF \(327KB\)](#)[View Record in Scopus](#)

29

S. Andaluz, et al.

Proteome profiles of thylakoid membrane and changes in response to iron deficiency

Photosynth. Res., 89 (2006), pp. 141-155

[CrossRefView Record in Scopus](#)

30

B. Naumann, et al.

Comparative quantitative proteomics to investigate the remodeling of bioenergetic pathways under iron deficiency in *Chlamydomonas reinhardtii*

Proteomics, 7 (2007), pp. 3964-3979

[CrossRefView Record in Scopus](#)

31

J.L. Moseley, et al.

Adaptation to Fe-deficiency requires remodeling of the photosynthetic apparatus

EMBO J., 21 (2002), pp. 6709-6720

[CrossRefView Record in Scopus](#)

32

S.I. Hsieh, et al.

The proteome of copper, iron, zinc and manganese micronutrient deficiency in *Chlamydomonas reinhardtii*

Mol. Cell. Biol., 12 (2013), pp. 65-86

[CrossRefView Record in Scopus](#)

33

A. Terauchi, et al.

Trophic status of *Chlamydomonas reinhardtii* influences the impact of iron deficiency on photosynthesis

Photosynth. Res., 105 (2010), pp. 39-49

[CrossRefView Record in Scopus](#)

34

N. Suzuki, et al.

ROS and redox signalling in the response of plants to abiotic stress

Plant Cell Environ., 35 (2012), pp. 259-270

[CrossRefView Record in Scopus](#)

35

B. Halliwell, J. Gutteridge

Oxygen toxicity, oxygen radicals, transition metals and disease

Biochem. J., 219 (1984), pp. 1-14

[CrossRefView Record in Scopus](#)

36

G. Vigani, et al.

Metabolic adjustment under Fe deficiency in roots of dicotyledonous plants

Y. Dincer (Ed.), *Iron Deficiency and its Complications*, Nova Science Publishers (2012), pp. 1-27

[View Record in Scopus](#)

37

J.F. Briat, et al.

Ferritins and iron storage in plants

Biochim. Biophys. Acta, 1800 (2010), pp. 806-814

[ArticlePDF \(974KB\)](#)[View Record in Scopus](#)

38

I. Coueè, et al.

Involvement of soluble sugars in reactive oxygen species balance and responses to oxidative stress in plants

J. Exp. Bot., 57 (2006), pp. 449-459

[CrossRefView Record in Scopus](#)

39

D.M. Rhoads, C.C. Subbaiah
Mitochondrial retrograde regulation in plants
Mitochondrion, 7 (2007), pp. 177-194
ArticlePDF (435KB)View Record in Scopus

40

R.E. Häusler, et al.
Chlororespiration and grana hyperstacking: how an Arabidopsis double mutant can survive despite defects in starch biosynthesis and daily carbon export from chloroplasts
Plant Physiol., 149 (2009), pp. 515-533
CrossRefView Record in Scopus

41

K. Apel, H. Hirt
Reactive oxygen species: metabolism, oxidative stress, and signal transduction
Annu. Rev. Plant Biol., 55 (2004), pp. 373-399
CrossRefView Record in Scopus

42

I.M. Møller, L.J. Sweetlove
ROS signalling – specificity is required
Trends Plant Sci., 15 (2010), pp. 370-374
ArticlePDF (229KB)View Record in Scopus

43

I.M. Møller, et al.
Oxidative modifications to cellular components in plants
Annu. Rev. Plant Biol., 58 (2007), pp. 459-481
CrossRefView Record in Scopus

44

L.J. Sweetlove, I.M. Møller
Oxidation of proteins in plants – mechanisms and consequences
Adv. Bot. Res., 52 (2009), pp. 1-23
ArticlePDF (337KB)View Record in Scopus

45

C.H. Foyer, G. Noctor
Redox sensing and signalling associated with reactive oxygen in chloroplasts, peroxisomes and mitochondria
Physiol. Plant., 119 (2003), pp. 355-364
CrossRefView Record in Scopus

46

N. Arnaud, et al.
An iron-induced nitric oxide burst precedes ubiquitin-dependent protein degradation for Arabidopsis AtFer1 ferritin gene expression
J. Biol. Chem., 281 (2006), pp. 23579-23588
CrossRefView Record in Scopus

47

M. Graziano, L. Lamattina
Nitric oxide and iron in plants: an emerging and converging story
Trends Plant Sci., 10 (2005), pp. 4-8
ArticlePDF (1MB)View Record in Scopus

48

C.W. Jin, et al.
Elevated carbon dioxide improves plant Fe nutrition through enhancing the Fe-deficiency-induced responses under Fe-limited conditions in tomato
Plant Physiol., 150 (2009), pp. 272-280
CrossRefView Record in Scopus

49

W.W. Chen, et al.

Nitric oxide acts downstream of auxin to trigger root ferric chelate reductase activity in response to iron deficiency in *Arabidopsis*

Plant Physiol., 154 (2010), pp. 810-819

[CrossRefView Record in Scopus](#)

50

C.W. Jin, et al.

NO synthase-generated NO acts downstream of auxin in regulating Fe deficiency-induced root branching that enhances Fe-deficiency tolerance in tomato plants

J. Exp. Bot., 62 (2011), pp. 3875-3884

[CrossRefView Record in Scopus](#)

51

K.J. Gupta, A.U. Igamberdiev

The anoxic plant mitochondrion as a nitrite: NO reductase

Mitochondrion, 11 (2011), pp. 537-543

[ArticlePDF \(513KB\)View Record in Scopus](#)

52

E. Gas, et al.

Hunting for plant nitric oxide synthase provides new evidence of a central role for plastids in nitric oxide metabolism

Plant Cell, 21 (2009), pp. 18-23

[CrossRefView Record in Scopus](#)

53

B. Touraine, et al.

GSH threshold requirement for NO-mediated expression of the *Arabidopsis* AtFer1 ferritin gene in response to iron

FEBS Lett., 586 (2012), pp. 880-883

[ArticlePDF \(591KB\)CrossRefView Record in Scopus](#)

54

J.D. Woodson, J. Chory

Coordination of gene expression between organellar and nuclear genomes

Nature, 9 (2008), pp. 383-395

[CrossRefView Record in Scopus](#)

55

T. Pfannschmidt

Plastidial retrograde signalling – a true ‘plastidial factor’ or just metabolite signatures?

Trends Plant Sci., 15 (2010), pp. 427-435

[ArticlePDF \(440KB\)View Record in Scopus](#)

56

S. Koussevitzky, et al.

Signals from chloroplasts converge to regulate nuclear gene expression

Science, 316 (2007), pp. 715-719

[CrossRefView Record in Scopus](#)

57

P.A. Salomé, et al.

Circadian clock adjustment to plant iron status depends on chloroplast and phytochrome function

EMBO J. (2013), 10.1038/emboj.2012.330

58

Y.Y. Chen, et al.

Iron is involved in maintenance of circadian period length in *Arabidopsis*

Plant Physiol. (2013), 10.1104/pp.112.212068

59

Y. Xiao, et al.

Retrograde signalling by the plastidial metabolite MEcPP regulates expression of nuclear stress-response genes

Cell, 149 (2012), pp. 1525-1535

[ArticlePDF \(1MB\)View Record in Scopus](#)

60

G.M. Estavillo, et al.

- Evidence for a SAL1-PAP chloroplast retrograde pathway that functions in drought and high light signalling in *Arabidopsis*
Plant Cell, 23 (2011), pp. 3992-4012
CrossRefView Record in Scopus
61
M. Seemann, et al.
Isoprenoid biosynthesis in chloroplasts via the methylerythritol phosphate pathway: the (E)-4-hydroxy-3-methylbut-2-enyl diphosphate synthase (GcpE) from *Arabidopsis thaliana* is a [4Fe-4S] protein J
Biol. Inorg. Chem., 10 (2005), pp. 131-137
CrossRefView Record in Scopus
62
M. Schwarzlander, et al.
The impact of impaired mitochondrial function on retrograde signalling: a meta-analysis of transcriptomic responses J
Exp. Bot., 63 (2012), pp. 1735-1750
CrossRefView Record in Scopus
63
D.G. Bernard, et al.
An allelic mutant series of ATM3 reveals its key role in the biogenesis of cytosolic iron-sulfur proteins in *Arabidopsis*
Plant Physiol., 151 (2009), pp. 590-602
CrossRefView Record in Scopus
64
J.C. Rutherford, et al.
A second iron-regulatory system in yeast independent of Aft1p
Proc. Natl. Acad. Sci. U.S.A., 98 (2001), pp. 14322-14327
CrossRefView Record in Scopus
65
P.L. Blaiseau, et al.
Aft2p, a novel iron-regulated transcription activator that modulates, with Aft1p, intracellular iron use and resistance to oxidative stress in yeast
J. Biol. Chem., 276 (2001), pp. 34221-34226
CrossRefView Record in Scopus
66
A. Kumánovics, et al.
Identification of FRA1 and FRA2 as genes involved in regulating the yeast iron regulon in response to decreased mitochondrial iron-sulfur cluster synthesis
J. Biol. Chem., 283 (2008), pp. 10276-10286
CrossRefView Record in Scopus
67
R. Ueta, et al.
Mechanism underlying the iron-dependent nuclear export of the ironresponsive transcription factor Aft1p in *Saccharomyces cerevisiae*
Mol. Biol. Cell, 18 (2007), pp. 2980-2990
CrossRefView Record in Scopus
68
H. Li, C.E. Outten
Monothiol CGFS glutaredoxins and BOLA-like proteins. [2Fe-2S] binding partners in iron homeostasis
Biochemistry, 51 (2012), pp. 4377-4389
CrossRefView Record in Scopus
69
S. Adinolfi, et al.
Bacterial frataxin CyaY is the gatekeeper of iron-sulfur cluster formation catalyzed by IscS
Nat. Struct. Mol. Biol., 16 (2009), pp. 390-396
CrossRefView Record in Scopus
70
A. Moreno-Cermeno, et al.
Frataxin depletion in yeast up-regulation of iron transport systems before affecting iron-sulfur enzyme activities

J. Biol. Chem., 53 (2010), pp. 41653-41664

[CrossRefView Record in Scopus](#)

71

J. Ihrig, et al.

Iron regulation through the back door: iron-dependent metabolite levels contribute to transcriptional adaptation to iron deprivation in *Saccharomyces cerevisiae*

Eukaryot. Cell, 9 (2010), pp. 460-471

[CrossRefView Record in Scopus](#)

72

S. Cesco, et al.

Release of plant-borne flavonoids into the rhizosphere and their role in plant nutrition

Plant Soil, 329 (2010), pp. 1-25

[CrossRefView Record in Scopus](#)

73

A. Higa, et al.

Iron deficiency induces changes in riboflavin secretion and the mitochondrial electron transport chain in hairy roots of *Hyoscyamus albus*

J. Plant Physiol., 167 (2010), pp. 870-878

[ArticlePDF \(916KB\)View Record in Scopus](#)

74

A. Vorwieger, et al.

Iron assimilation and transcription factor controlled synthesis of riboflavin in plants

Planta, 226 (2007), pp. 147-158

[CrossRefView Record in Scopus](#)

75

G. Vert, et al.

Dual regulation of the Arabidopsis high-affinity root iron uptake system by local and long-distance signals

Plant Physiol., 132 (2003), pp. 796-804

[CrossRefView Record in Scopus](#)

76

M. John, et al.

Cell signaling by oligosaccharides

Trends Plant Sci., 2 (1997), pp. 111-115

[ArticlePDF \(794KB\)View Record in Scopus](#)

77

C. Curie, J.F. Briat

Iron transport and signalling in plants

Annu. Rev. Plant Biol., 54 (2003), pp. 183-206

[CrossRefView Record in Scopus](#)

78

R. Ivanov, et al.

Fitting into the harsh reality: regulation of iron-deficiency responses in dicotyledonous plants

Mol. Plant, 5 (2011), pp. 27-42

[CrossRefView Record in Scopus](#)

79

D. Duy, et al.

PIC1, an ancient permease in Arabidopsis chloroplasts, mediates iron transport

Plant Cell, 19 (2007), pp. 986-1006

[CrossRefView Record in Scopus](#)

80

D. Duy, et al.

The chloroplast permease PIC1 regulates plant growth and development by directing homeostasis and transport of iron

Plant Physiol., 155 (2011), pp. 1709-1722

[CrossRefView Record in Scopus](#)

81

K. Bashir, et al.

Identification and characterization of the major mitochondrial Fe transporter in rice

Plant Signal. Behav., 6 (2011), pp. 1591-1593

[CrossRefView Record in Scopus](#)

82

S.A. Kim, et al.

Localization of iron in Arabidopsis seed requires the vacuolar membrane transporter VIT1

Science, 314 (2006), pp. 1295-1298

[CrossRefView Record in Scopus](#)

83

K. Ravet, et al.

Post-translational regulation of AtFER2 ferritin in response to intracellular iron trafficking during fruit development in Arabidopsis

Mol. Plant, 2 (2009), pp. 1095-1106

[ArticlePDF \(570KB\)](#)[CrossRefView Record in Scopus](#)

84

D. Leister

Retrograde signalling in plants: from simple to complex scenarios

Front. Plant Sci., 3 (2012), p. 135

85

J.D. Woodson, J. Chory

Organelle signalling: how stressed chloroplasts communicate with the nucleus

Curr. Biol., 22 (2012), pp. R690-R692

[ArticlePDF \(110KB\)](#)[View Record in Scopus](#)