Biochimica et Biophysica Acta 1850 (2015) 1479–1496

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbagen

Review

# Involvement of thiol-based mechanisms in plant development ☆

Nicolas Rouhier [a,b], Delphine Cerveau [c,d,e], Jérémy Couturier [a,b], Jean-Philippe Reichheld [f,g], Pascal Rey [c,d,e,*]

[a] Université de Lorraine, Interactions Arbres—Microorganismes, UMR1136, F-54500 Vandoeuvre-lès-Nancy, France
[b] INRA, Interactions Arbres—Microorganismes, UMR1136, F-54280 Champenoux, France
[c] CEA, DSV, IBEB, Laboratoire d'Ecophysiologie Moléculaire des Plantes, Saint-Paul-lez-Durance F-13108, France
[d] CNRS, UMR 7265 Biologie Végétale & Microbiologie Environnementale, Saint-Paul-lez-Durance F-13108, France
[e] Aix-Marseille Université, Marseille F-13284, France
[f] Laboratoire Génome et Développement des Plantes, Université Perpignan Via Domitia, F-66860 Perpignan, France
[g] Laboratoire Génome et Développement des Plantes, CNRS, F-66860 Perpignan, France

ARTICLE INFO

*Article history:*
Received 2 October 2014
Received in revised form 8 January 2015
Accepted 10 January 2015
Available online 9 February 2015

*Keywords:*
Glutathione
Glutaredoxin
Plant development
Reductase
Thiol group
Thioredoxin

ABSTRACT

*Background:* Increasing knowledge has been recently gained regarding the redox regulation of plant developmental stages.
*Scope of view:* The current state of knowledge concerning the involvement of glutathione, glutaredoxins and thioredoxins in plant development is reviewed.
*Major conclusions:* The control of the thiol redox status is mainly ensured by glutathione (GSH), a cysteine-containing tripeptide and by reductases sharing redox-active cysteines, glutaredoxins (GRXs) and thioredoxins (TRXs). Indeed, thiol groups present in many regulatory proteins and metabolic enzymes are prone to oxidation, ultimately leading to post-translational modifications such as disulfide bond formation or glutathionylation. This review focuses on the involvement of GSH, GRXs and TRXs in plant development. Recent studies showed that the proper functioning of root and shoot apical meristems depends on glutathione content and redox status, which regulate, among others, cell cycle and hormone-related processes. A critical role of GRXs in the formation of floral organs has been uncovered, likely through the redox regulation of TGA transcription factor activity. TRXs fulfill many functions in plant development via the regulation of embryo formation, the control of cell-to-cell communication, the mobilization of seed reserves, the biogenesis of chloroplastic structures, the metabolism of carbon and the maintenance of cell redox homeostasis. This review also highlights the tight relationships between thiols, hormones and carbon metabolism, allowing a proper development of plants in relation with the varying environment and the energy availability.
*General significance:* GSH, GRXs and TRXs play key roles during the whole plant developmental cycle via their antioxidant functions and the redox-regulation of signaling pathways. This article is part of a Special Issue entitled Redox regulation of differentiation and de-differentiation.



## 1. Introduction

Thiols, the sulfur analogues of alcohols, are organo-sulfur compounds that contain a carbon-bonded sulfhydryl group (R–SH). Thiols are readily prone to oxidation especially in a basic environment since thiol deprotonation leads to the formation of thiolate ($R\text{-}S^-$), a nucleophilic form sensitive to oxidation. In the presence of reactive oxygen species (ROS), such as hydrogen peroxide, oxidation leads to the formation of a sulfenic acid form (R-SOH) that can be further oxidized to sulfinic and sulfonic forms ($R\text{-}SO_2H$ and $R\text{-}SO_3H$, respectively) [1]. Of note, thiol groups can also be oxidized by reactive nitrogen species resulting for instance in S-nitrosylation (R-SNO) [2]. In other respects, when an oxidized cysteine residue is brought near a reduced cysteine residue, it generates a cystine unit with a disulfide bond (R-S-S-R′) that can alter protein folding, contributing to tertiary structure if the cysteines belong to the same polypeptide or to quaternary structure if they belong to different polypeptides. Owing to the physico-chemical properties of thiol groups and to their capacity to bind metals, proteins having these reactive cysteines play key roles in biology. They contribute for instance to ROS detoxification, to the control of cell redox homeostasis or to signaling transduction pathways in particular via redox post-translational modifications.

In most living organisms, thiol groups are mainly present in the sulfur-containing amino acid, cysteine, which is present in most proteins and is also a component of glutathione, a γ-Glu-Cys-Gly tripeptide existing either in a reduced form (GSH) or in oxidized forms, such as glutathione disulfide (GSSG) or nitrosoglutathione

☆ This article is part of a Special Issue entitled Redox regulation of differentiation and de-differentiation.

* Corresponding author at: CEA, DSV, IBEB, Laboratoire d'Ecophysiologie Moléculaire des Plantes, Saint-Paul-lez-Durance F-13108, France. Tel.: +33 442254776.
*E-mail address:* pascal.rey@cea.fr (P. Rey).

http://dx.doi.org/10.1016/j.bbagen.2015.01.023
0304-4165/

(GSNO). In plants, glutathione is present in most tissues with subcellular concentrations in the millimolar range [3,4] and is considered together with ascorbate as a major redox buffer [3,5]. Two enzymes: γ-glutamyl-cysteine ligase or γ-glutamyl-cysteine synthase (γ-GCS or GSH1) and glutathione synthetase (GS or GSH2) ensure the production of glutathione at the expense of ATP. The reduction of GSSG is catalyzed by glutathione reductase (GR), a NADPH-dependent flavoprotein [5]. The redox status of protein cysteinyl residues is controlled by two main types of enzymes termed thioredoxins (TRXs) and glutaredoxins (GRXs). Many of them, particularly TRXs, display disulfide reductase activity, and some are able to reduce sulfenic acid forms. GRXs are more specifically involved in the deglutathionylation of proteins reversibly modified by the formation of a mixed disulfide bond between glutathione and a cysteine. However, whereas it is firmly established that all GRXs from class I possess deglutathionylation activities, it is less clear whether GRXs belonging to classes II and III possess such activity [6]. Both TRXs and GRXs function via a catalytic site formed at least by a redox active cysteine which is generally separated in the primary sequence by two variable residues from another cysteine (often referred to as resolving cysteine) in most TRXs and in about half of GRXs or from a serine in the half remaining GRXs [6,7]. A WCGPC amino acid signature is generally quite well conserved among TRXs whereas a greater variability is found among GRXs being often of the CPYC, CPFC, CGFS or CCxC/S type. To obtain a complete view of the active site sequences and current classification of GRXs and TRXs, we invite the reader to refer to the following papers [8,9]. At the structural level, TRXs and GRXs adopt a specific TRX-fold that is usually characterized by a 4-stranded parallel beta-sheet core enclosed by 4 to 5 alpha-helices, and that is shared by several other protein families. In plant cells, the regeneration of oxidized TRXs and GRXs is generally achieved via distinct pathways (Fig. 1). For the large majority, GRXs use reduced glutathione as an electron donor but for example, a chloroplastic class II Grx from *Chlamydomonas reinhardtii* was shown to be recycled by the ferredoxin-Trx reductase [7,10]. The reduction of TRXs is more complex and further depends on their subcellular localization. Nuclear, cytosolic and mitochondrial TRXs are usually reduced by NADPH-thioredoxin reductases (NTR), enzymes with FAD cofactors [11] whereas chloroplastic TRXs are reduced in a light-dependent manner by a ferredoxin-dependent thioredoxin reductase (FTR), an iron-sulfur (Fe–S) enzyme composed of two different subunits and which converts the electron signal coming from photosystem I and relayed by ferredoxins into a thiol-reducing cascade [12]. Atypical reduction modes exist for some TRXs. A poplar TRX h4 and its plant orthologs are regenerated by the GSH/GRX system owing to the presence of an additional cysteine in the N-terminal region [13–15]. Some Trx-like and Trx-lilium can be recycled by GSH in a manner analogous to GRXs [16].

Until the last years, glutathione was mainly thought to act as a redox buffer in various processes like responses to environmental stress, plant–pathogen interactions and detoxification of xenobiotics and heavy metals [5]. Plant TRXs have been long presumed to mainly regulate photosynthesis and carbon metabolism enzymes [17] or to participate in the mobilization of seed reserves [18]. They have been also characterized as important actors in the responses of plants to oxidative stress [19]. The information concerning the sequence characteristics, the biochemical functions or the expression patterns of GRXs lag behind, since they have been identified later in plants compared with TRXs [7]. Although some have been reported quite early to play roles in flower development [20,21] and later in stress responses [22,23], the physiological roles of many GRXs remain elusive.

A role for TRXs and GRXs as key determinants in plant growth and development has only been recently uncovered together with the involvement of several ROS like NO or $H_2O_2$. This is in fact not surprising considering the capacity of the latter molecules to modify protein thiol groups. The cellular redox homeostasis is varying during specific developmental stages or in response to changing environmental conditions. It has thus to be tightly regulated and among the various types of effectors, thiol-containing components form an important signaling network. An additional layer of new information concerning the involvement of redox regulated circuits into developmental processes is the complex interplays that have been unveiled between glutathione in particular and several hormones. In this review, we describe the current knowledge and recent advances concerning the roles of glutathione and of disulfide reductases, GRXs and TRXs, in the control of development in relation to ROS and hormones.

## 2. Glutathione is essential for plant development

### 2.1. Direct involvement of glutathione in developmental stages of plants

Glutathione is synthesized in two steps catalyzed by the gamma-glutamyl cysteine synthase (GSH1) and the glutathione synthase (GSH2). In higher plants, GSH1 is exclusively located in plastids whereas GSH2 is dual-targeted to plastids and cytosol [24,25]. The crucial role of glutathione for plant development was first demonstrated by the characterization of different mutants in the biosynthesis pathway (Table 1). Unless GSH is added to the growth medium, Arabidopsis

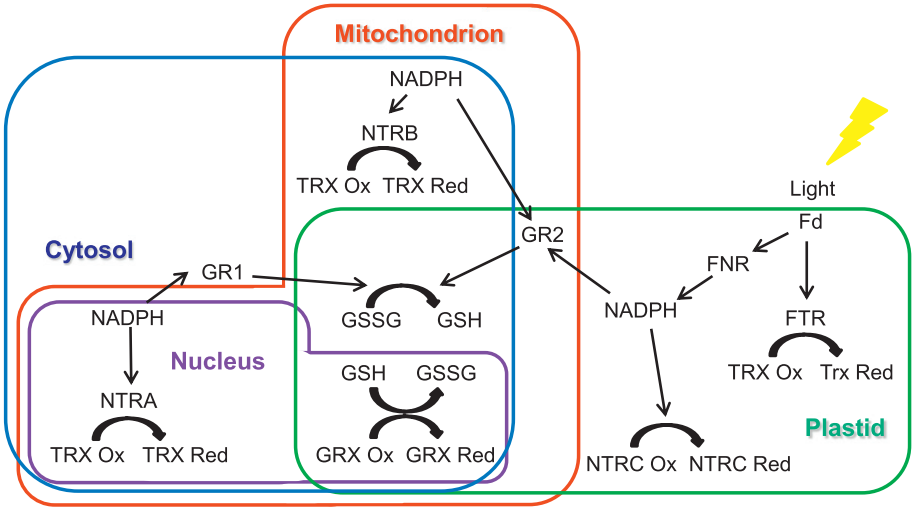


**Fig. 1.** Reduction pathways of the thiol-containing compounds, glutathione, glutaredoxins and thioredoxins in the main subcellular compartments of plant cells. GSH and GSSG, reduced and oxidized glutathione, respectively; GR, glutathione reductase (2 types, 1 and 2); GRX Ox and Red, oxidized and reduced glutaredoxin, respectively; TRX Ox and Red, oxidized and reduced thioredoxin, respectively; NTR: NADPH thioredoxin reductase (3 types: A, B and C); Fd, ferredoxin; FNR, ferredoxin $NADP^+$ reductase; FTR, ferredoxin thioredoxin reductase.

knock-out lines for *AtGSH1* are embryo-lethal [26] whereas those for *AtGSH2* exhibit normal embryogenesis, but are lethal at the seedling stage [25]. In fact, the *gsh2* mutant accumulates high levels of gamma-glutamyl-cysteine (γ-EC), but this precursor cannot replace glutathione [25]. Interestingly, in some legumes, homoglutathione, a glutathione derivative, is formed owing to the existence of a homoglutathione synthetase which catalyzes the addition of an alanine instead of a glycine to γ-EC. In these species, both GSH and homoglutathione are important for the formation of nodules, specialized organs formed during the symbiotic association of these plants with rhizobial bacteria and allowing atmospheric nitrogen fixation [27].

The presence of GSH in all subcellular compartments, coupled to the fact that GSH1 should only be found in plastids regardless the plant species considered, pointed to the need to transport γ-EC and/or GSH from the chloroplast to the cytosol and then to transport GSH to all compartments. A family of three transporters in *Arabidopsis thaliana*, namely chloroquine resistant (CRT)-like transporters (CLTs), was shown to transport γ-EC and GSH across the chloroplast envelope [28]. In accordance with the observed glutathione deficiency in roots, a triple *clt1 clt2 clt3* Arabidopsis mutant exhibits sensitivity to cadmium and to pathogen attack. The latter effect would result from the requirement of glutathione or its γ-EC precursor for pathogenesis-related (*PR*) gene induction [28]. Moreover, the fact that this triple mutant is not lethal suggests that an additional transporter for GSH or its precursor should exist.

In Arabidopsis, an allelic series of five *gsh1* mutants displays various glutathione content, comprised between 3% and 50% of the wild-type level [29–33]. The *rml1* allele exhibits the lowest glutathione level (3%) and the most severe developmental defects. In this mutant, the post-embryonic development of roots, but not of shoots, is impaired [29,34]. Combining the *rml1* mutation with disruption in the two genes encoding the NADPH-thioredoxin reductases A and B (*AtNTRA* and *AtNTRB*) showed that the shoot apical meristem function is in fact controlled both by glutathione and the TRX system [35]. Three mutants with a higher glutathione content, *pad2-1* (22%), *cad2-1* (20%) and *zir1* (15%), grow normally under control conditions but are more sensitive to biotic stress, cadmium and zinc treatments respectively [31–33,36]. In the *pad2-1* mutant, pollen germination and early elongation of the pollen tube are affected, as observed by depletion of the glutathione level using buthionine sulfoximine (BSO) [37]. Incidentally, pollen fertility is also perturbed in the double *ntra ntrb* mutant and in mutants affected in both TRX and glutathione reduction pathways [35,38]. In the *cad2-1* mutant, the primary root growth phenotype is mild, but a pronounced effect on lateral root development is visible [39]. The mutant *regulator of AtAPX2 1-1* (*rax1-1*) has about 40–50% of the wild-type foliar glutathione level and was identified by constitutive expression of the photo-oxidative stress-inducible ascorbate peroxidase 2 [31]. The *rax1-1* mutant does not exhibit any developmental phenotype but under photo-oxidative stress conditions, the expression of a wide set of defense-related genes is altered. The difference in glutathione content in all these mutants is explained by the existence of distinct point mutations which lead to a change of different amino acid residues near the GSH1 active site [40]. Overall, the comparison of the growth perturbations in all these mutants indicates that a basal glutathione level (between 3% and 15%) is required for proper meristem functioning and plant development in optimal conditions (Fig. 2).

Other reports support the idea that, in addition to the global glutathione amount, the glutathione redox status is also essential for plant development [28,41–43]. Whereas a cytosolic/peroxisomal glutathione reductase *gr1* mutant is aphenotypic, a knock-out Arabidopsis mutant for the chloroplastic/mitochondrial AtGR2 is embryo-lethal [44]. The study of *miao*, a weak allele of the plastidial glutathione reductase AtGR2, indicates that this isoform is essential for root growth and root apical meristem maintenance through both auxin/PLETHORA (PLT)-dependent and auxin/PLT-independent redox signaling pathways [43]. A mutant for the *AtATM3* gene coding for a mitochondrial ABC transporter, which connects the mitochondrial Fe–S cluster assembly machinery to the biosynthesis of nuclear/cytosolic Fe–S proteins, accumulates high level of GSSG in the mitochondria and exhibits a perturbed root development [45].

### 2.2. Requirement of glutathione for cell cycle progression

The role of glutathione in cell proliferation has been largely documented [46–48]. High levels of glutathione are generally associated with active cell proliferation. For example, actively dividing root meristematic cells possess high levels of glutathione which is moreover mostly in the reduced form whereas the quiescent center maintains low levels of glutathione in a highly oxidized state [48,50,51]. Depleting the glutathione pool by BSO was shown to block the cell cycle progression in the G1 phase, which might explain why *gsh1* and *gsh2* mutants are unable to establish post-embryonic root or shoot meristems [29,35,49]. Both in mammals and plants, the measurement of GSH pools using a fluorescent probe, 5-chloromethylfluorescein diacetate (CMFDA), suggested that glutathione is massively recruited in the nucleus early in the cell proliferation cycle [52,53]. On the contrary, depletion of nuclear glutathione was shown to impair cell proliferation [54]. The mechanism that would allow the nuclear sequestration of GSH is not known in plants but similarly to mammals, it could involve proteins resembling the anti-apoptotic factor Bcl-2 [53]. It was proposed that the nuclear glutathione recruitment in G1 promotes oxidation in the cytosol and enhances glutathione biosynthesis [55]. In Medicago, a plant having both glutathione and homoglutathione, distinct roles in the regulation of the cell cycle have been attributed to these molecules. Glutathione is associated with meristematic cells, promoting cell cycle activation and induction of somatic embryogenesis, whereas homoglutathione is associated with differentiated cells and embryo proliferation [56].

### 2.3. The interplay between glutathione and hormones

Glutathione is a key actor of the interplay between redox and plant hormone signaling pathways. It is involved in particular in plant defense responses by modulating salicylic acid (SA) and jasmonic acid signaling pathways [57–59], and specific GRXs may constitute mediators of this response [60]. For instance, accumulation of glutathione is triggered by fungal elicitors, pathogen infection and exogenous application of the defense-related hormone SA [61–63]. The best characterized example is the involvement of GSNO in conjunction with the cytosolic AtTRX h5 in the regulation of the SA-induced systemic acquired resistance pathway by modulating the oligomerization state and subcellular localization of the AtNPR1 (Non-expresser of PR genes 1) transcriptional regulator [63–67]. We invite the reader to refer to the following review [68] for more details on all these aspects.

A link between GSH and abscisic acid (ABA) was evident from the observation that the glutathione content in guard cells decreases along with ABA-induced stomatal closure and that genetic or pharmacological depletion of glutathione enhances ABA-induced stomatal closure [69]. The exact contribution of GSH for this process is not known. However, since $H_2O_2$ and NO represent key signaling molecules, it is very likely that important redox changes should occur and that GSH, alone or in conjunction with GRXs, participates in the signaling cascades for example by regulating protein redox modifications.

The best documented link between GSH and hormones in developmental processes concerns auxin. Indeed, several lines of evidence suggest that glutathione and auxin metabolisms are intimately linked, at least in roots a very convenient model for studying cell elongation and differentiation, owing in particular to the simplicity to perform cell imaging in this organ. Auxin accumulation in root stem cells is clearly dependent on their redox status [47,51,70]. Upon BSO treatment of Wt seedlings and in several glutathione biosynthesis mutants, the auxin maximum in the root tips is impaired and the auxin transport is

**Table 1**
Developmental defects associated to the knock-out or knock-down of genes involved in thiol homeostasis.
Among the hundredth of *GRX* and *TRX* genes, mutants for only a few genes have been studied so far. However, the key role of the GSH/GRX and TRX systems for development appears clear since mutants deficient for GSH biosynthesis (GSH1 & GSH2) or reduction (GRs) or for TRX reduction (NTRA & B, FTR) generally have either strong or lethal phenotypes. Developmental defects associated to the whole allelic series for *AtGSH1* genes (*pad2-1*, *cad2-1*, *zir1*, *rax1-1*, *rml1*) are not described here but in the text.

| Protein names | Plant species | Gene locus | Type of mutants | Subcellular localization | Mutant phenotype(s) | References |
|---|---|---|---|---|---|---|
| Synthesis and reduction of glutathione and its derivatives | | | | | | |
| GSH1 | *Arabidopsis thaliana* | At4g23100 | T-DNA insertion, EMS mutagenized | Chloroplast | Knock-out mutant embryo-lethal, strong allele shows severely impaired post-embryonic growth and weak alleles defects in pollen and root development | [26,29–33] |
| GSH2 | *Arabidopsis thaliana* | At5g27380 | T-DNA insertion | Chloroplast/cytosol | Seedling-lethal | [25] |
| GR1 | *Arabidopsis thaliana* | At3g24170 | T-DNA insertion | Cytosol/peroxisome | No growth phenotype | [38] |
| GR2 | *Arabidopsis thaliana* | At3g54660 | T-DNA insertion, EMS mutagenized | Mitochondrion/chloroplast | Knock-out mutant embryo-lethal, weak alleles have defects in root growth and in root apical meristem maintenance | [43,44] |
| GSNOR | *Arabidopsis thaliana* | At5g43940 | T-DNA insertion, EMS mutagenized, antisense transgenic plants | Nucleocytoplasmic | Reduced root length, defects in stem and trichome branching, reduced fertility, loss of heat acclimation loss of apical dominance, reduced hypocotyl elongation, decreased silique size and seed production | [228–231] |
| Glutaredoxins | | | | | | |
| GRXC1/C2 | *Arabidopsis thaliana* | At5g63030/At5g40370 | T-DNA insertion | Nucleocytoplasmic | Embryo-lethal | [104] |
| GRXS17 | *Arabidopsis thaliana* | At4g04950 | T-DNA insertion and RNAi lines | Nucleocytoplasmic | Hypersensitivity to elevated temperature and long photoperiod altered auxin perception | [22, 106] |
| GRXS13 | *Arabidopsis thaliana* | At1g03850 | RNAi lines | Nucleocytoplasmic | Reduced plant growth | [119] |
| ROXY1 | *Arabidopsis thaliana* | At3g02000 | T-DNA insertion | Nucleocytoplasmic | Impaired petal primordia initiation & petal morphogenesis | [20] |
| ROXY2 | *Arabidopsis thaliana* | At5g14070 | T-DNA insertion | Nucleocytoplasmic | Impaired anther development | [21] |
| ROXY4 | *Arabidopsis thaliana* | At3g62950 | T-DNA insertion | Nucleocytoplasmic | Impaired anther development | [107] |
| MIL1 | *Oryza sativa* | Os07g05630 | Spontaneous mutant | Nucleocytoplasmic | Defective meiosis | [108] |
| MSCA1 | *Zea mays* | CAX52135 | EMS mutagenized | Nucleocytoplasmic | Impaired anther development | [109–111] |
| Thioredoxins and targets | | | | | | |
| NTRa/b | *Arabidopsis thaliana* | At2g17420/At4g35460 | T-DNA insertion | Mitochondrion/cytosol | Wrinkled seed, slower plant growth, reduced pollen fitness | [35] |
| FTR catalytic subunit | *Arabidopsis thaliana* | At2g04700 | Virus induced gene silencing | Chloroplast | Abnorrmal chloroplast development | [146] |
| NTRc | *Arabidopsis thaliana* | At2g41680 | T-DNA insertion | Chloroplast | Retarded growth of shoots and roots, defects in lateral root formation | [168–170] |
| TRX m3 | *Arabidopsis thaliana* | At2g15570 | EMS mutagenized, Ds transposon | Chloroplast | Embryo-lethal, impaired meristem development | [128] |
| TRX m | *Oryza sativa* | Os12g08730 | RNAi lines | Chloroplast | Abnorrmal chloroplast development | [159] |
| TRXs m1, 2, 4 | *Arabidopsis thaliana* | At1g03680/At4g03520/ At3g15360 | Virus induced gene silencing | Chloroplast | Pale-green leaves and reduced stability of PSII | [91] |
| TRX m | *Pisum sativum* | CAA45098/CAA53900 | Virus induced gene silencing | Chloroplast | Pale-green leaves, reduced pigment content and lower photosynthetic capacity | [160] |
| TRX z | *Arabidopsis thaliana* | At3g06730 | T-DNA insertion | Chloroplast | Albino phenotype | [15,147] |
| TRX z | *Nicotiana benthamiana* | AY500242 | Virus-induced gene silencing | Chloroplast | Albino phenotype | [147] |
| TRX h9 | *Arabidopsis thaliana* | At3g08710 | T-DNA insertion | Plasma membrane | Dwarf plants with short roots and small yellowish leaves | [15] |
| THL1 and THL2 | *Brassica napus* | AAB53694/AAB53695 | Antisense transgenic plants | Cytosol | Self-incompatibility rejection response with reduced pollen adhesion, germination and pollen tube growth | [137] |
| NRX1 | *Arabidopsis thaliana* | At1g60420 | T-DNA insertion | Nucleocytoplasmic | Reduced pollen fertility | [133] |
| AtECB1 | *Arabidopsis thaliana* | At4g28590 | Ds transposon & T-DNA insertion | Chloroplast | Albino phenotype, impaired chloroplast development | [152] |
| HCF164 | *Arabidopsis thaliana* | At4g37200 | T-DNA insertion | Chloroplast | Impaired accumulation of cytochrome b6f complex subunits | [155] |
| GPX5 | *Arabidopsis thaliana* | At3g63080 | Ds transposon insertion | Plasma membrane | Arrested embryo development | [211] |
| GPX3 | *Oryza sativa* | Os02g44500 | RNAi lines | Mitochondrion | Reduced root and shoot development | [208] |
| GPX1/GPX7 | *Arabidopsis* | At2g25080/At4g31870 | RNAi lines | Chloroplast | Altered leaf cells and chloroplast | [210] |

**Table 1** (*continued*)

| Protein names | Plant species | Gene locus | Type of mutants | Subcellular localization | Mutant phenotype(s) | References |
|---|---|---|---|---|---|---|
| MSRB2 | *thaliana Capsicum annuum* | EF144171 | Virus induced gene silencing | Chloroplast | morphology Reduced shoot development | [216] |

affected [39,71]. Microarray analyses performed on these plants have revealed that the transcriptional reprogramming observed upon glutathione depletion is very similar to the effects linked to auxin homeostasis deregulation [71,72]. In particular, glutathione depletion by BSO has major effects on auxin distribution in the roots and it perturbs the expression of genes coding for proteins involved in auxin metabolism and transport (*PIN* genes) [39,71]. The possible link between auxin signaling and glutathione was reinforced by the observation that the auxin-resistant Arabidopsis mutants, *axr1* and *axr3* are less sensitive to BSO treatment than wild-type plants [71]. Although the mechanism underlying these observations is not yet known, it is likely that glutathione redox imbalance triggers oxidation of components of the auxin signaling pathway. Indeed, the TRANSPORT INHIBITOR RESPONSE 1 (AtTIR1) auxin receptor has been shown to be sensitive to NO-mediated oxidation through S-nitrosylation of a critical Cys residue [73]. Moreover, the use of both chemical treatments and Arabidopsis mutants with altered NO levels has also demonstrated that high levels of NO reduce auxin transport and response via a PIN1-dependent mechanism [74]. A role for ROS in auxin metabolism has also been proposed. A ROS-dependent transcriptional modulation of auxin signaling components mediates the responses to oxidative stress [75]. Moreover, works on the Arabidopsis *cat2* mutant which accumulates $H_2O_2$ compared to wild type plants showed that $H_2O_2$ modulates auxin metabolism [76,77]. Finally, the $H_2O_2$-responsive UDP-glucosyltransferase UGT74E2 which is involved in the regulation of auxin homeostasis through the conversion of auxin indole-3-butyric acid (IBA) to IAA was shown to modulate plant architecture in Arabidopsis [78]. The existence of interplays between glutathione and other phytohormones is likely, although not well-documented at the present time. For instance, a role for glutathione in the regulation of root architecture has been found very recently in relation with the metabolism of strigolactones [79].

### *2.4. Indirect roles of glutathione via metabolic processes and responses to environmental constraints*

Previous studies conducted in yeast indicate that glutathione may be primarily required for two major cellular processes, the protein folding in the endoplasmic reticulum and the regulation of iron metabolism, notably the assembly of extra-mitochondrial Fe–S enzymes [80,81]. Indeed, the glutathione requirement for the latter process comes from the fact that a glutathione derivative could provide the sulfur exported from mitochondria and necessary for the building of Fe–S clusters in cytosolic/nuclear proteins via the cytosolic iron/sulfur cluster assembly (CIA) machinery [82]. Owing to their capacity to bind [2Fe–2S] clusters using GSH molecules, GRXs may well serve as relays for this cellular process [83]. Several indispensable DNA-modifying enzymes such as helicases or endonucleases require the binding of Fe–S clusters for their folding and/or activity [82]. This explains why in plants, yeasts or animals, mutants for components involved in the maturation of Fe–S clusters are often lethal [82,84]. Hence, while it is difficult to examine this point without performing a thorough molecular analysis of conditional glutathione-deficient mutant plants for example, the observed lethality of *gsh1* and *gsh2* mutants in Arabidopsis may be primarily due to a defect in the biogenesis of Fe–S clusters, leading to non-functional cell structures.

The key role of glutathione in many developmental pathways underlies the fact that it also serves as a substrate for several enzymes such as GRXs, glutathione S-transferases (GSTs) or dehydroascorbate reductases (DHARs). The role of specific GRXs in different aspects of plant development is documented and will be detailed in the Section 3.2. In addition to their known functions in plant defense against oxidants and xenobiotics, some GSTs have developmental functions. For example, GSTU17 was proposed to participate in light signaling in Arabidopsis through phytochrome A-dependent regulated expression. GSTU17 also participates in various aspects of seedling development, including hypocotyl elongation and root development by modulating the pool of glutathione in an auxin-dependent manner [85,86]. DHARs are responsible for regenerating ascorbate from the oxidized dehydroascorbate form. Consequently, they regulate the cellular ascorbate redox state, which affects the tolerance to environmental constraints leading to variations in ROS content. A role of DHAR in plant development was also demonstrated in transgenic tobacco lines down-regulated for DHAR activity. In these plants, growth and leaf aging were affected as a consequence of higher foliar ROS level and impaired photosynthetic activity during leaf development [87].

All these data highlight the crucial roles of glutathione in almost all steps of plant development. The redox buffer capacity of this compound is likely responsible for the high adaptability of plants to environmental constraints. Although the exact contribution of glutathione in plant development is still to decipher, it is likely mediated through posttranslational modifications of key Cys residues in target proteins. Finding the targets of glutathionylation involved in plant development will constitute the next challenge to specify the functions of glutathione. Several proteomic approaches dedicated to the isolation and identification of glutathionylated proteins have already been used and developed in photosynthetic organisms [88]. In other respects, the presumed role of glutathione for the maturation of Fe–S clusters suggests its participation in key pathways of plant development processes for instance via the biogenesis and functioning of cell structures.

## 3. Roles of TRXs and GRXs in plant development

Compared to other organisms, which most often display one to three *TRX* and *GRX* genes fulfilling various physiological functions, terrestrial plants are characterized by a remarkable diversity since there are *ca.* 50 genes encoding TRX and TRX-like proteins and *ca.* 30 genes encoding GRXs [8,9,89]. Concerning GRXs, this high number is specifically due to the considerable increase during evolution of a specific class referred to as class III [8]. Indeed, the class III GRXs are restricted to land plants, the moss *Physcomitrella patens* containing only 2 isoforms compared to 21 isoforms present in *A. thaliana*. Whether this diversity is associated with functional redundancy or specialization is a longstanding question since the physiological role of most plant TRXs and GRXs was indefinite until the last years. But, in accordance with the observation that several plant TRXs and GRXs are expressed in very specific cell or tissue types, recent data gained notably from genetic approaches revealed that some plant TRXs and GRXs fulfill unique and specialized functions related to responses to stress or development. For the case of the specific expansion of class III GRXs in land plants, evolution analyses suggested that subfunctionalization and subsequent neofunctionalization facilitated their conservation [90]. Functional redundancy and virtual

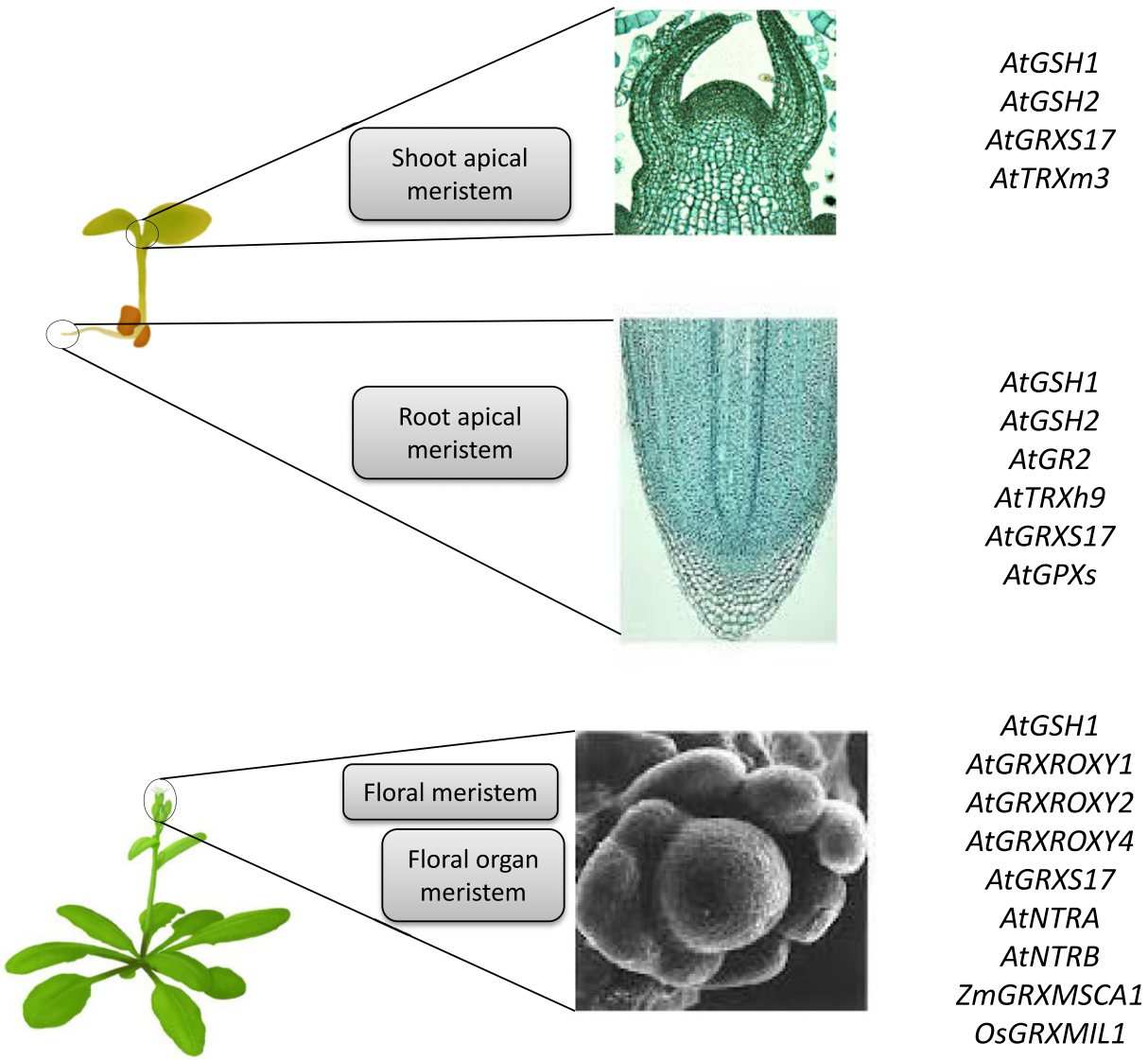


**Fig. 2.** Involvement of genes participating in glutathione synthesis and reduction or encoding thiol-dependent reductases in the maintenance of plant meristems. The roles of these genes have been established on the basis of functional genetic approaches. The relevant references are in Table 1.

interchangeability have been highlighted for members sharing similar sequence and biochemical characteristics and belonging to large subgroups such as plastidial TRXs m [91] and class III GRXs [92]. But, in that case, it is also likely that distinct spatial and temporal patterns of expression result in unique physiological functions.

### *3.1. Expression patterns of TRX and GRX in organs, tissues or at specific developmental stages*

Based on Northern blots, semi-quantitative RT-PCR, promoter–GUS fusions and in silico analysis of Arabidopsis transcriptomic data, the 9 genes coding for cytosolic or mitochondrial TRX h isoforms have been reported to exhibit very distinct expression patterns [93,94]. *AtTRX h7* is specifically expressed in root vascular tissues and *AtTRX h8* only in ovaries. Others show a low expression in all plant tissues, but exhibit high amounts of transcripts in the stele and mature pollen (*AtTRX h1*) or in unicellular pollen and dry seed (*AtTRX h4*). This is also true for other Arabidopsis *TRX* gene types. Just to cite an example, the gene coding for the plastidial ACHT3 (atypical Cys His-rich TRX) is highly expressed in pollen [95,96]. But at the present time, the precise functions of most of these TRXs remain unknown.

In other plant species, similar conclusions have been drawn regarding the expression pattern of *TRX* genes. In pea, the *PsTRX h1* transcript is detected in most organs by real-time PCR, with higher levels in leaves and flowers [97] and high expression of the gene was reported in pollen grains and stigmata using the *GUS* reporter system [97]. Many plant *TRXs h* genes are also characterized by specific expression in seeds particularly during germination as shown in pea [98], and in the model legume *Medicago truncatula* [99]. In wheat (*Triticum aestivum*), the expression of three *TaTRX h* genes is induced during the desiccation phase, whereas only one of them, *TaTRX h1* is expressed after imbibition [100]. In legumes, the formation of root nodules requires specific signaling pathways and coordinated gene expression between symbiotic partners. In soybean, a *TRX h* gene is specifically expressed in particular root cells during the nodulation process and in infected cells [101]. The encoded TRX has been proposed to participate in the regulation of redox homeostasis for proper nodule development. The requirement of TRXs in this process has been extended by Alkhalfioui et al. [102] who

isolated two novel TRXs specific to Legumes termed TRXs s. Based on quantitative RT-PCR and Western analyses showing specific and high expression in nodules, these two isoforms have been proposed to participate in the symbiotic interaction. On the basis of biochemical data, plastidial TRXs f and m are presumed to regulate the photosynthetic metabolism [17], and accordingly, immunolocalization indicated the presence of both proteins in leaf photosynthetic cells in pea plants [103]. Further, this method revealed distinct expression patterns particularly in non-photosynthetic organs, since PsTRX m is highly abundant in vascular tissues of leaves, stems and roots whereas PsTRXf is less abundant and localized in specific cells at proximity of xylem vessels and vascular cambium [103].

With regard to *AtGRX* genes, analysis of microarray data revealed that those belonging to the two first classes (At*GRXC1* to *AtGRXC5* and *AtGRXS12* for class I and *AtGRXS14* to *AtGRXS17* for class II [7]) are expressed in most organs of Arabidopsis plants, except in the male gametophyte where only *AtGRXC2* and *AtGRXS16* transcripts are detected [94]. *AtGRXC1* and *AtGRXC3* genes are more specifically expressed in the root columella and in vascular tissues, respectively, whereas *AtGRXS14* exhibits high expression in all photosynthetic tissues [94]. On the contrary, many *AtGRX* members from class III, termed ROXY, display more specific expression profiles. For example, microarray data indicate that transcripts of *AtROXY6* and of *AtROXY8-16* are specifically present in root procambium and those of *AtROXY2*, *AtROXY4*, *AtROXY7*, *AtROXY8* and *AtROXY12–15* in the phloem mainly in root. However, transcriptomics data need to be validated using other methods to better delineate expression patterns. For instance, in the case of *AtROXY1*, microarray data indicate the presence of transcripts mainly in the shoot apical meristem [94] whereas investigations performed using RT-PCR and in situ hybridization reveal a quite different pattern, with a high transcript level in roots, siliques and inflorescences [20]. Within inflorescences, *AtROXY1* is specifically expressed in the apex and in floral organ primordia [20].

### 3.2. Involvement of GRXs and TRXs in plant developmental stages

#### 3.2.1. Roles of GRXs in the development of plants

Only a few studies unveiled the roles of class I and class II GRXs for plant development. Whereas single *A. thaliana* insertion mutants for *AtGRXC1* and *AtGRXC2* have no visible phenotype under standard growth conditions, the double mutant is lethal at an early stage after pollination (Table 1) [104]. In accordance to their similar biochemical properties and subcellular localizations [94,105], it points to the possible redundant function between both proteins. It is worth noting that *At*GRXC1 orthologs are only found in dicots. Hence, in other plants, GRXC2 may be the only representative ensuring this function. The other example described so far for which developmental defects were observed is for GRXS17, a class II GRX. The Arabidopsis knock-out mutant for this gene presents pleiotropic phenotypes which are likely linked to the deregulation of temperature and photoperiod signaling pathways, notably in the meristem [22,106]. Indeed, in conditions of long photoperiod or high temperature, *grxS17* plants show abnormal apical meristem, elongated leaves and impaired flowering. A complete growth arrest is observed when both environmental conditions are combined [106]. At the root level, a decrease in the primary root growth is visible at 22 °C and exacerbated at 28 °C [22]. In the latter condition, auxin sensitivity and polar auxin transport as well as cell proliferation and/or cell cycle control are impaired in *grxS17* plants. Hence, in line with the observed connexions between glutathione and auxin and with their requirement for proper cell division, GRXS17 appears to be an important integrator for redox and auxin signaling pathways, although the associated molecular mechanisms and in particular the target proteins of this GRX are not yet elucidated.

*3.2.1.1. Involvement of class III GRXs in the development of floral organs.* The well-described involvement of class III GRXs in plant development and more particularly in floral development comes from genetic studies performed primarily in *Arabidopsis thaliana* and *Oryza sativa* (Table 1, Fig. 2). In *A. thaliana*, the flower organization comprises four concentric whorls corresponding to different organs with four sepals in the first whorl, four petals in the second whorl, six stamens in the third whorl and two carpels in the fourth whorl. A role of class III GRX isoforms in floral development was initially demonstrated in Arabidopsis from the identification of the *roxy1* mutant, which exhibits a reduced number of petal primordia and abnormalities in petal morphogenesis [20]. The inability of a gene version where the catalytic cysteine was replaced by a serine to complement the *roxy1* mutant suggested that the oxidoreductase activity of the protein is crucial for this function [20]. Finally, the authors also demonstrated that AtROXY1 also participates in the negative regulation of *AGAMOUS* gene expression in the first and second whorls, this protein functioning in the development of floral organs of third and fourth whorls [20].

This initial study was then followed by the characterization of *AtROXY2*, the closest homolog of *AtROXY1*. Contrary to the *roxy1* mutant, the *roxy2* mutant does not display any perturbation in floral development. However, in addition to having a reduced number of petals, the double mutant *roxy1 roxy2* is sterile and does not produce pollen [21]. Further analysis demonstrated that both proteins are involved in early adaxial and abaxial anther and tapetum development but with a distinct timing. Expressing *AtROXY2* under the control of *AtROXY1* promoter fully rescues the *roxy1* mutant phenotype [21]. Hence, based on this result and on overlapping expression of *AtROXY1* and *AtROXY2* genes in anther primordia, it has been proposed that both proteins act redundantly to secure the production of pollen and thus plant fertility, notably in stress conditions [21].

Besides these two GRXs, another class III GRX isoform, ROXY4, plays a role in floral development. Initially identified as an immediate target up-regulated by the DELLA protein RGA, AtROXY4 was suggested to participate in gibberellin signaling and floral organ development. Indeed, it is highly expressed in sepals and plants overexpressing *AtROXY4* displayed undeveloped petals and stamens and male sterility due to non-dehiscent anthers [107]. Interestingly, the involvement of some class III GRXs in floral development is conserved between monocots and dicots since two rice ROXY1 homologs, *OsROXY1* and *OsROXY2*, fully rescue the floral phenotype of Arabidopsis *roxy1* mutant [92]. In addition, the expression patterns of the two rice genes in floral development are similar to those of *AtROXY1* and *AtROXY2* in Arabidopsis [92]. The last evidence of a role of class III GRXs in floral development comes from the study of mechanisms regulating the transition to meiosis in rice [108]. The authors isolated the *mil1* mutant that does not produce microspores in anthers and is thus male sterile, but female fertile. Although *OsMIL1* codes for a class III GRX isoform, the phylogenetic analyses indicate that it is not the ortholog of Arabidopsis *ROXY1* and *ROXY2*. The molecular and cytological analyses of the rice *mil1* mutant showed that the meiotic entry of sporogenous cells is defective resulting in the formation of somatic cells instead of microspores [108]. Therefore, the authors suggested that male meiosis initiation is subject to redox regulation and that OsMIL1 plays a key role in this process. Accordingly, the mutation in maize of a class III *GRX* gene named *ZmMSCA1* leads to male sterile lines [109–111]. One of these studies further highlights that redox components like ZmMSCA1, in connection with the $O_2$ concentration level, are important for the switch from somatic to germline cell formation [110].

*3.2.1.2. Proposed roles for GRXs in plant development.* The role of class III GRXs in the control of floral development notably relies on their capacity to interact with TGA transcription factors as shown primarily in Arabidopsis [101]. A GRX/TGA interaction was confirmed in other species such as rice where MIL1 was shown to interact with TGA1 [108]. A nuclear interaction between *A. thaliana* ROXY1 and PAN, a

TGA member acting as a repressor of petal formation, was demonstrated [112,113]. Note that the nuclear localization of AtROXY1 is indispensable for its function in petal development. The question of a redox control of PAN by ROXY1 was investigated. A mutated version of AtPAN, where the cysteine 340 is replaced by a serine, is not able to complement the *pan* Arabidopsis mutant [112]. Altogether, these results suggest that AtROXY1 may regulate AtPAN activity by modulating its redox state. However, although the active site signature of these GRXs (CCxx) contains the strictly conserved pair of cysteines, the difficulties encountered to express and purify recombinant proteins prevent to definitely conclude about the requirement of an oxidoreductase activity for this function [114].

In Arabidopsis, the phenotypic comparison of a double *roxy1 pan* mutant with the corresponding single mutants showed that double *roxy1 pan* and *pan* mutants have 5 petals whereas *roxy1* mutant has only 2.5 petals [112]. It indicates that PAN is epistatic to AtROXY1 and thus both proteins act in the same regulatory pathway controlling petal primordia initiation. Furthermore, AtROXY1 acts independently of PAN during later petal morphogenesis. Hence, in addition to PAN, ROXY1 may modulate the activity of other TGA factors displaying similar expression patterns, like TGA9 and TGA10 [112,115]. Accordingly, the double *tga9 tga10* Arabidopsis mutant exhibits defects in male gametogenesis similar to those of the double *roxy1 roxy2* mutant, and a nuclear interaction between AtROXY1 and AtTGA9 and AtTGA10 was demonstrated [115]. Hence, it seems that class III GRXs can interact with multiple TGA factors and that these interactions depend on the presence of specific motifs. For instance, two motifs, L**LL and ALWL, present in the C-terminal part of ROXY1 are important for its function in floral development [112,116]. Interestingly, the L**LL motif is crucial for the interaction between ROXY1 and PAN or TGA3 whereas the ALWL motif, though dispensable for TGA interaction, is required for petal development [112,116]. A refined L***L motif was shown to be important for the interaction of many Arabidopsis class III GRXs with TGAs [116,117].

From these data and given the high number of class III GRX isoforms in plants, it is tempting to speculate that the uncharacterized members are involved in the development of organs other than flowers. Supporting this view, the analysis of *O. sativa* and *A. thaliana* transcriptomic data indicated that some of these *GRX* genes present specific expression patterns [94,118]. From an evolutionary point of view, it is remarkable to note that the increasing number of class III GRXs between the moss *P. patens* and higher plants is concomitant with the development of more complex structures. Another factor that likely explains this huge expansion is the fact that several class III GRXs, like AtGRXS13, are also involved in pathogen responses and more generally in stress responses [23,60,117,119]. In other respects, *A. thaliana* lines silenced for *AtGRXS13* already display a reduced growth compared to that of wild type plants in optimal conditions [119].

### 3.2.2. Roles of TRXs in the development of plants

*3.2.2.1. Potential TRX targets participating in plant development.* Since their discovery almost forty years ago, plant TRXs have been shown to participate in the regulation of photosynthetic metabolism, the mobilization of seed reserves and the responses to environmental constraints. These functions are most generally achieved via their disulfide reductase activity, which enables them to control the redox status of target proteins and thus to switch on or off their activity. Intriguingly, although a high number of potential TRX partners has been identified in the last years [120], the knowledge regarding the TRX targets directly involved in developmental processes are still very poor. By analogy to the control of TGA factors by class III GRXs, the activity of transcription factors controlling plant development might also be subjected to TRX-mediated post-translational modifications. Among possible effects, redox modifications can lead to activation or deactivation via conformational change or nucleo-cytosolic shuttling as observed for the AtNPR1 transcription factor [66]. While the redox-regulation of gene transcription is well established in bacteria, fungi and mammals for some time, similar insights have only been recently gained in the plant kingdom [121]. For instance, the DNA-binding activity of maize R2R3 MYB-domain proteins is redox-regulated in vitro via two cysteines able to form a disulfide bridge [122]. Similarly, a cysteine is required for the binding activity of an Arabidopsis leucine zipper (bZIP) type transcription factor, named AtbZIP16, and the redox status of this factor has been proposed to control its activity in relation with environmental conditions [123]. In other respects, homeodomain-leucine zipper (HD-Zip) class III proteins constitute a complex family of transcription factors controlling the development of embryo and meristem in plants [124,125] and sharing two conserved cysteines that form a disulfide bridge [126]. Most interestingly, the in vitro DNA-binding capacity of the recombinant or native form of Athb-9 HD-Zip III factor is activated by TRX [126]. Similarly, class I TCP (Teosinte branched1-Cycloidea Proliferating cell factor) transcription factors regulate plant developmental processes. Most of them possess a conserved cysteine involved in the formation of an intermolecular dimer, which is inactive and can be reactivated by the TRX system [127]. Overall, these data give strong credence to a role of cytosolic/nuclear TRXs in the regulation of the expression of key developmental genes through the modulation of transcription factor activity.

*3.2.2.2. Roles of TRXs in vegetative and reproductive development.* Unexpectedly, when taking into consideration the key role of NTRs in the electron supply to nuclear, cytosolic and mitochondrial TRXs, an Arabidopsis mutant line deficient for both *AtNTR* genes (*A* and *B*) is viable and displays slower growth and lower pollen fitness [35]. But, AtTRX h3 is still partially reduced in this mutant, indicating the participation of an alternative mechanism in the reduction of TRXs. Interestingly, when crossing this double mutant with others impaired in glutathione biosynthesis (*rml1* or *cad2*), very severe developmental phenotypes were observed such as complete growth arrest in the case of *ntra ntrb rml1* [35] or naked floral stems and loss of apical dominance in *ntra ntrb cad2* plants [39]. The phenotype of *ntra ntrb cad2* resembles that of Arabidopsis mutants impaired in auxin metabolism and accordingly, the auxin transport capacity and cellular level are altered in the triple mutant. These reports show redundancy between NTR and glutathione-dependent systems in the reduction of TRXs and in the control of plant development. Further, they reveal the participation of redoxins in auxin homeostasis possibly through the redox control of auxin-related proteins.

In the last years, two Arabidopsis T-DNA mutants knock-out for the expression of very distinct types of TRXs have been reported to develop dramatically altered phenotypes with regard to meristem functioning, possibly due to defects in cell-to-cell communication mechanisms (Table 1, Fig. 2). Thus, Benitez-Alfonso et al. isolated an Arabidopsis seedling-lethal mutant due to impaired meristem maintenance resulting from disrupted unloading from the phloem into the meristem [128]. This mutant turned out to be defective in the *AtTRX m3* gene. This gene coding for a plastidial TRX expressed in non-green plastids of meristems and organ primordia plays thus a critical role in development. Accordingly, ectopic expression of this TRX leads to delayed flowering and senescence. AtTRX m3 has been proposed to control callose deposition and symplastic permeability in a redox manner, thus modifying plasmodesmata communication.

Regarding *AtTRX h* genes, an Arabidopsis mutant knock-out for *AtTRX h9* expression is dwarf and develops short roots and pale leaves [15]. In fact, this protein possesses an N-terminal extension with one highly conserved glycine at the second position and one cysteine at the fourth position which are required for membrane binding and intercellular movement, possibly via myristoylation and palmitoylation, respectively. This plasma membrane associated TRX is able to move from cell to cell in root layers, suggesting its

participation in intercellular communication mechanisms underlying developmental steps. In rice, the OsTRX h1 isoform also plays a key developmental function since plants deficient in this protein which is secreted into the extracellular space are dwarf and have a reduced tiller number [129]. As these modified plants also display increased level of $H_2O_2$, OsTRX h1 has been proposed to regulate redox homeostasis in the apoplast, but its precise role in the control of plant development is still mysterious. Finally, based on expression profiles, other relatively unexpected roles for TRXs h in plant development can be presumed. For instance, the mechanisms underlying gravitropism, which is essential for appropriate plant growth, are still very obscure. Interestingly, *TRX h* expression is triggered in poplar following gravistimulation and numerous TRX targets are differentially expressed after this stimulus [130], revealing a possible role of TRX-based mechanisms in gravitropism-linked signaling pathways located in the endodermal cells.

*3.2.2.3. Roles of TRXs in gamete formation and fertilization.* As mentioned in the Section 3.1 and thoroughly reviewed by Traverso et al. [131], numerous TRX types are expressed in plant sexual organs. However, their precise roles in plant reproduction are still elusive in most cases. Some genetic studies performed in the last years showed the participation of specific TRXs like h-type TRXs and nucleoredoxins (NRXs). A role for at least one AtTRXs h in the proper development of male gametes is evident from the demonstration that a double *ntra ntrb* mutant, deficient in the reduction of cytosolic TRXs, displays altered pollen fitness [35]. Both Arabidopsis *NRX* genes are expressed in reproductive organs. Based on increased pollen sterility in *nrx1* knock-out plants [132,133], AtNRX1 has been proposed to participate in the establishment of pollen fertility [133]. A dual nuclear/cytosolic localization was found for AtNRX1, but the targeted protein(s) by this TRX is (are) unknown.

One of the best documented examples concerning the participation of TRXs in plant reproduction is self-incompatibility. This mechanism which prevents self-fertilization is based on the interaction or not of specific male and female *S*-determinants, which differ among plant families. In self-incompatible species, the pollen is recognized and rejected by stigma papillae. In Brassicaceae, self-incompatibility notably involves an S-locus receptor kinase (SRK) on the female side, which mediates pollen recognition following binding to the *S*-locus Cys-rich (SCR) protein from the male side. In the case of self-pollen, the binding activates a signaling pathway leading to rejection through SRK kinase activity. Interestingly, this phosphorylation step can be blocked by an h-type TRX termed THL1 in *Brassica oleracea* [134–136]. This TRX thus acts as a negative regulator of SRK in the absence of self-incompatible pollen. In other respects, Haffani et al. [137] reported that antisense suppression of *BnTHL1* and *BnTHL2* expression leads to a low but constitutive level of self-pollen rejection in a normally compatible *Brassica napus* cultivar. From these data, a model in which THL-type TRXs are required for successful pollination in self-incompatible Brassicaceae species has been proposed. But this model has been recently questioned by Yamamoto and Nasrallah [138], who reported no change in SRK activation in Arabidopsis lines expressing the SRK-SCR system from the self-incompatible species, *Arabidopsis lyrata*, and deficient for the expression of Arabidopsis THL homologues. Further, these authors also reported that AlSRK is not redox-regulated by AtTRX proteins. Of note, the participation of TRXs h in pollen compatibility mechanisms has been hypothesized in other plant families. In Solanaceae, a *Nicotiana alata* TRX h isoform is secreted into the extracellular matrix of style transmitting tract and is able to interact in vitro with *S*-RNase, a determinant of pollen recognition in the pistil in this plant family [139]. This TRX has been proposed to fulfill a role in S-RNase-based self-incompatibility. In *Phalaris coerulescens*, a plant of the Poaceae family, a pollen S-protein contains a TRX h domain displaying reductase activity [140,141]. Very interestingly, a self-fertile line of this species is characterized by mutation in the PcTRX h domain leading to a decreased disulfide reductase activity [142]. Altogether, these results indicate that the redox control of self-incompatibility seems quite widespread in plants.

*3.2.3. A model for thiol-based control of plant morphogenesis*

The available data, gained from genetics studies show that various plant GRX and TRX isoforms fulfill critical roles in key developmental steps, such as cell fate determination in meristems, and reveal the requirement of disulfide reductases for the proper progress of organ morphogenesis. But, the precise mechanisms underlying these roles are still elusive. They could involve post-translational redox modification of partners acting in signaling transduction pathways or in control of gene expression, as proposed for class III GRXs with TGA factors. The next challenge to give further credence to this hypothesis will consist in identifying these actors. Of note, such a model has been validated for TRXs in plant immunity and pathogen responses, which involve NPR1, a master regulator in the expression of defense genes. The activity of this regulator, which is able to interact with and to activate TGA factors, is modulated via salicylic acid and a redox control of its quaternary structure and subcellular localization by both nitric oxide and AtTRX h5 [66,143].

*3.3. Involvement of TRXs in plant development through the biogenesis of cell structures*

In plant chloroplasts, FTR is the primary reducer of TRXs linking together with ferredoxins the photosynthetic electron transfer chain to several biosynthetic and assimilation pathways [144]. This protein is composed of a catalytic subunit (FTRc) and a variable subunit (FTRv). While Arabidopsis mutants deficient for one of the variable subunits, AtFTRv, display no phenotype except an increased sensitivity to oxidative treatments [145], lines silenced for the expression of the catalytic subunit exhibit reduced growth and a sectored chlorotic leaf phenotype [146]. In these silenced lines, the expression of genes controlled by the plastid-encoded RNA polymerase (PEP) complex is strongly reduced supporting the hypothesis that FTR participates in the regulation of the PEP function in the early stages of chloroplast development [146]. A *TRX z* deficient Arabidopsis mutant line has also a decreased expression of PEP-dependent plastidial genes and an *albino* phenotype characteristic of impaired chloroplast development [15,147]. As a consequence, TRX z appears as a serious candidate for PEP regulation, since the poplar isoform is reduced by FTR [148]. Although the Arabidopsis TRX z is not reduced by FTR, its dimeric form is reduced by other plastidial TRXs [149]. The question of the requirement of TRX z redox activity in PEP functioning is still unanswered because of the existence of contradictory results. Whereas AtTRX z was initially shown to interact with and reduce two fructokinase-like (FLN) proteins, other subunits of the PEP complex, complementation studies of *trx* z mutants with redox inactive (i.e. cysteine mutated) AtTRX z forms suggested that the redox activity is dispensable for PEP functioning under standard conditions [147,150]. From the latter observation it was proposed that AtTRX z and AtFLN rather play a structural function in the complex formation and/or maintenance. Among others, one possible explanation to reconcile all results is that the presence of the redox inactive AtTRX z is sufficient to recruit other redox factors that by-pass the lack of redox active TRX. Of note, other components like AtECB1/MRL7, which has been shown to interact with TRX z, and AtTACS are required for PEP function [151–153]. Although AtECB1 does not possess the typical CxxC active site signature, it contains three cysteines, displays a C-terminal domain with TRX-like fold and exhibits disulfide reductase activity towards insulin [152]. Hence, the exact function of all these proteins is still to be determined to delineate their roles in the expression of plastidial genes.

The biogenesis of cytochrome $b_6f$ complex is another redox-regulated process that is crucial for plant development. It requires the

transfer of reducing equivalents from the stroma to the thylakoid lumen. The electrons are likely provided by an m-type TRX present in the stromal side, then transferred to the thylakoid-anchored AtCCDA protein before reaching AtHCF164 (High Chlorophyll Fluorescence 164), which is located in the thylakoid lumen [154]. In fact, AtHCF164, which displays disulfide reductase activity, was the first plant TRX-like protein identified [155]. A T-DNA mutant deficient in this gene is not able to grow photoautotrophically and its reproductive development is indeed abolished due to impaired accumulation of cytochrome $b_6f$ complex [155]. The loss-of-function Arabidopsis mutants for the *AtCCDA* gene show a similar though less pronounced phenotype, i.e. pale green leaves, severely reduced growth and altered reproductive development [156]. These phenotypes clearly originate from disrupted photosynthesis and defects in cytochrome $b_6f$ maturation as indicated by the measured high chlorophyll fluorescence level. It has been proposed that AtHCF164 and AtCCDA participate to the delivery of reducing power to keep cysteines of the apocytochrome in a reduced state prior to heme ligation [156].

Plastidial TRXs f and m have been the first plant TRXs characterized and in vitro biochemical investigations revealed that these TRX types (2 and 4 isoforms, respectively, in Arabidopsis) regulate in a light-dependent manner the activity of enzymes participating in photosynthesis and carbon metabolism [157,158]. A notable exception is AtTRX m3 that only shows poor activation properties toward these enzymes, which can be understood considering its specific involvement in meristem maintenance [128]. However, the precise physiological function of all other plastidial TRXs was not clear until these last years. A severely impaired phenotype, i.e. reduced leaf and root growth, pale-green leaves and low grain yield, was reported for rice plants knock-down for Os*TRX m* expression [159]. In this case, the disturbance of plastidial redox homeostasis leads to abnormal chloroplast development and to modified pigment contents. Consistently, the simultaneous silencing of *AtTRXs m1*, *m2* and *m4* genes in Arabidopsis plants also caused reduced growth, pale-green leaves and elevated ROS levels [91]. Based on the interaction of the three AtTRX m isoforms with numerous PSII subunits and on the altered redox status of PSII core subunits in plants deficient for the three TRXs, AtTRXs m have been proposed to participate in PSII biogenesis and thus to control the formation of photosynthetic complexes. This critical role of TRXs m has been confirmed in pea plants. Whereas the silencing of *PsTRX f* expression does not lead to noticeable change in growth and development, the simultaneous silencing of *PsTRXs f* and *m* genes results in pale-green leaves, altered redox homeostasis and impaired photosynthesis [160]. This phenotype has been attributed at least partly to defects in the chlorophyll biosynthesis pathway, notably due to impaired redox regulation of the magnesium chelatase CHLI subunit. Interestingly, transplastomic *Nicotiana tabacum* plants overexpressing at high level one *NtTRX m* gene, compared to others over-expressing one *NtTRX f* gene, specifically exhibit substantially reduced growth, lower chlorophyll content and altered glutathione and ascorbate pools [161]. Taken collectively, these data indicate that plastidial TRXs m, in addition to their role in the regulation of carbon metabolism enzymes, participate in crucial steps of chloroplast development via the control of the biogenesis of photosynthetic complexes.

Intriguingly, at the present time, the participation of TRXs in plant developmental processes through the biogenesis of cell structures has been established exclusively for various types of plastidial isoforms. Such a physiological function could be specific to the biogenesis of photosynthetic membranes in chloroplasts, in which the conversion of light energy to chemical energy, notably reducing power, is performed. Whether this type of function is fulfilled in other cell compartments like nucleus and mitochondria remains to be established. But it is worth mentioning that in these organelles much less TRX isoforms are present compared to plastids [89].

### *3.4. Involvement of TRXs in plant development through the regulation of metabolism*

Mature seeds develop in an oxidative cellular context, particularly during the last maturation phases (desiccation) and germination, as shown by the high level of protein oxidation measured at these stages [162,163]. The control of the redox status of proteins appears thus crucial for proper seed development and germination. As reported in various plant species, some TRXs h isoforms are highly expressed at various stages of seed maturation and during germination [98–100]. These TRXs are able to reduce in vitro various types of metabolic and storage seed proteins such as α-amylases, trypsin inhibitors, gliadins and glutenins [18,164]. In agreement with these expression and biochemical data, overexpression of wheat TRX h in barley is associated with accelerated germination and triggers the activities of pullulanase, a starch-debranching enzyme and of α-amylase [165,166]. These data reveal a critical role of TRXs in the germination process through the control of the mobilization of reserves. The TRX control is likely achieved via their disulfide reductase activity, which allows the activation of metabolic enzymes and the reduction of storage proteins thereby rendering them soluble and available for providing the energy necessary for seedling development.

Another TRX playing an important role in the control of metabolic pathways is the plastidial NADPH-dependent thioredoxin reductase C (NTRC). This unique, plant specific, enzyme formed by an NTR domain fused to a C-terminal TRX domain was described in Arabidopsis and rice [167-168]. An Arabidopsis mutant knock-out for *AtNTRC* shows reduced growth compared to wild type and is more sensitive to methyl viologen, salt and drought stresses [168]. Further investigations revealed that short-day conditions induce severe metabolic and developmental disturbance in *ntrc* plants. This phenotype is linked to reduced chlorophyll and anthocyanin contents, to low carbon assimilation and to increases in transpiration and photorespiration [169,170]. Overexpression of *AtNTRC* allows increasing the leaf size and the biomass yield in both short- and long-day conditions confirming that NTRC is a key actor for plant development [171]. The low amounts of chlorophyll in the *ntrc* mutant are linked to reduced synthesis of 5-aminolevulinic acid, the chlorophyll precursor, and decreased MgP methyltransferase activity [172]. Arabidopsis *ntrc* plants also show reduced root development, with slower growth rate and fewer lateral roots than wild type, revealing an important role of AtNTRC in non-photosynthetic tissues [170]. Surprisingly, all these phenotype traits are photoperiod-dependent since they are less severe under long-day conditions. The role of AtNTRC in the regulation of starch synthesis could partly explain this phenotype. Indeed, *ntrc* plants show decreased redox-activation of ADP-Glucose pyrophosphorylase (AGPase) and are not able to synthesize enough starch during the light period. In short-day conditions, NTRC appears as an important redox regulator of starch synthesis and plastid metabolism in photosynthetic and non-photosynthetic plastids [173, 174]. Thus, these metabolic changes ultimately affect developmental processes, which are linked to the energy availability in cells. But, one cannot exclude that other biochemical properties underlie the NTRC function since the Arabidopsis protein can act as a chaperone with foldase and holdase activities [175]. These multiple functions are linked to modifications in the protein quaternary structure: low-molecular-weight complexes exhibit disulfide reductase activity whereas high-molecular-weight complexes preferentially show chaperone activity. The transition from low to high molecular weight complexes is stimulated by heat shock [175]. NTRC may not be the only TRX contributing to the regulation of starch synthesis. Indeed, the TRX f type has been reported to control starch synthesis based on observations performed in Arabidopsis *TRX f1* knock-out lines, which display no obvious growth phenotype, but decreased starch accumulation probably resulting from reduced light-activation of AGPase [176]. In agreement, the starch content in transplastomic

tobacco lines overexpressing *NtTRX f* is increased [177], but their growth is somewhat reduced compared to wild type [161].

Another more or less direct contribution of plastidial TRXs in development could come from their relationship to brassinosteroids (BRs). These are plant steroid hormones having key roles in the control of plant growth and development [178], and also in the responses to stresses through regulation of redox homeostasis and $CO_2$ assimilation [179]. Interestingly, the expression of genes encoding various TRX types has been recently reported to be regulated by these hormones. BR-deficient tomato plants are dwarf and display decreased transcript levels of *TRXs f*, *m* and *x* whereas exogenous application of BRs triggers the expression of these genes [180]. The authors proposed that plastidial TRXs participate in the BRs-initiated signaling transduction pathways regulating metabolism in relation with the plant developmental program.

To conclude, the regulation of metabolism by TRXs in connection with hormonal and environmental signals appears essential for proper plant development. This role is very likely achieved through the regulation of the activity of metabolic enzymes via redox post-translational modification. Accordingly, many enzymes involved in the metabolism of sugars, starch, amino acids and lipids have been characterized as regulated by TRXs or as potential targets of these reductases [120]. The fine redox control of their activity might allow supplying the particular metabolic needs required for the optimal progress of the various developmental stages.

### 3.5. Involvement of TRXs in plant development through antioxidant functions

In their natural environment, plants are continuously exposed to varying climatic conditions and changes in the light excitation pressure. As a consequence, their growth and development, which rely on a genetic program, are substantially influenced by environmental factors that condition the energy availability and modify the cellular redox homeostasis, particularly in the chloroplast where photosynthesis takes place. To prevent situations of oxidative stress, plants developed a complex and large array of antioxidant compounds notably low-molecular-weight molecules such as glutathione and ascorbate [3] and enzymes such as superoxide dismutases, catalases and peroxidases [181]. The participation of some TRX targets in this regulation network has been uncovered relatively recently, thanks to proteomic methods mainly based on affinity trapping or co-immunoprecipitation using mutant TRX forms and on thiol labeling using fluorescent or radioactive probes [120].

#### 3.5.1. TRXs supply electrons to antioxidant enzymes

Several plant TRXs participate to the responses to oxidative stress conditions resulting from environmental constraints [19]. This antioxidant role is notably achieved through the supply of reducing power to peroxiredoxins (PRXs) and methionine sulfoxide reductases (MSRs), enzymes reducing organic peroxides [182] and repairing oxidized proteins [183], respectively, thanks to redox-active cysteines. CDSP32 (chloroplastic drought-induced stress protein of 32 kDa) is a good example as it provides electrons to PRXs and MSRs as shown by in vitro and *ex vivo* experiments [184–186]. This is a double module TRX first isolated in potato plants subjected to water deficit [187]. Interestingly, the StCDSP32 protein abundance substantially decreases with leaf age in well-watered plants and this developmental pattern of expression is abolished in water stress conditions, which result in decreased and increased TRX abundance in young and adult leaves, respectively [188]. The higher CDSP32 expression in young leaves is very likely related to the fact that these leaves, in which the photosynthetic machinery is not fully functional and prone to photo-damage, are characterized compared to adult and senescing leaves by more efficient antioxidant defense mechanisms even in the absence of environmental constraints [189,190]. As a consequence, the TRXs like CDSP32 having an antioxidant function might have an indirect role for the good progress of plant development through the preservation of cell structures at very specific stages such as young developing leaves.

The plastidial y-type TRXs are also able to reduce PRXs and MSRs in biochemical assays [158,191–193]. Based on the expression patterns in Arabidopsis of the two *AtTRX y* genes, *y1* and *y2*, they could fulfill antioxidant functions at specific stages of plant development since *AtTRX y1* is expressed in non-photosynthetic organs such as seeds whereas *AtTRX y2* is mostly expressed in leaves [191]. The TRX y function in these organs could be related to their capacity to reduce MSRs. Indeed, Arabidopsis mutant lines deficient in *AtTRX y2* have a decreased leaf MSR capacity and display reduced growth and pale leaves when cultivated in high-light and long-day conditions [194]. Of note, MSRs likely play a key role in the preservation of seed longevity in *M. truncatula* and *A. thaliana* [195]. It is thus tempting to propose a specific role for the TRX y1 isoform in the maintenance of the protein repair capacity and in the preservation of longevity in seeds.

#### 3.5.2. Roles of thiol peroxidases in plant development via the control of peroxide levels

It is quite well documented in animals that $H_2O_2$ is important for several cellular processes including cell proliferation [196]. In plants, only a few examples demonstrated a role of ROS, including $H_2O_2$, in development. In *Arabidopsis thaliana* roots, it was shown that the transition from proliferation to differentiation depends on gradients of superoxide and $H_2O_2$ with superoxide accumulating in dividing cells and $H_2O_2$ in cells from the elongation zone [197]. Other studies showed that these two ROS species are also critical for root elongation/expansion and root hair differentiation [198,199]. Considering the roles played by peroxides, $H_2O_2$ in particular, for the development and signaling, the roles of enzymes locally controlling their concentrations are likely to be crucial. Among them, PRXs and glutathione peroxidases (GPXs) are non-heme thiol peroxidases that catalyze the reduction of $H_2O_2$ or organic hydroperoxides to water and alcohols via a catalytic cysteine residue [182,192]. The substrate reduction results in the oxidation of the thiol group of the so-called peroxidatic cysteine in a sulfenic acid form. The regeneration can occur via direct reduction by TRXs but this is not the majority of cases. In general, the sulfenic acid reacts with another cysteine residue coming either from the PRX or from GSH and leading to the formation of a disulfide bond that is subsequently reduced by TRXs or GRXs.

In *A. thaliana*, PRXs are encoded by nine nuclear genes and classified in four groups, 1-Cys PRX, PRXII, PRXQ and 2-Cys PRX, based on the number and position of redox-active cysteines [200]. 1-Cys PRX is specifically expressed during late seed development in the aleurone and in embryo cells, and is involved more particularly in the inhibition of germination under environmental constraint conditions. 1-Cys PRX has been proposed to be both a dormancy regulator and an antioxidant based on data gained in Arabidopsis and in cereals. It has been hypothesized that overoxidation of this PRX would constitute a signal to inhibit germination under oxidative stress conditions [201–204]. Among the Arabidopsis PRXs, AtPRXII-F is the only one targeted to mitochondria. Under optimal growth conditions, an Arabidopsis mutant knock-out for *PRXII-F* has no specific phenotype whereas it exhibits strong inhibition of root growth under stress conditions [205,206].

A relationship has been reported between the GPX activity and the inhibition of root tip growth induced by various abiotic stresses like heavy metals, high salt and $H_2O_2$ treatment in barley [207]. Recent investigations in rice provided evidence about the involvement of mitochondrial OsGPX3 in plant development [208]. OsGPX3 is the predominant GPX isoform expressed in roots of rice. Transgenic lines deficient for *OsGPX3* show shorter roots concomitant to increased $H_2O_2$ production in these organs. These data suggest that OsGPX3 contributes to maintaining adequate $H_2O_2$ levels for root growth [209]. In *A. thaliana*, 8 GPXs are present and named AtGPX1 to AtGPX8. The importance of GPXs for root architecture has been

analyzed in this species by characterizing T-DNA insertion lines. Five lines knock-out for the expression of *AtGPX1*, *AtGPX4*, *AtGPX6*, *AtGPX7* or *AtGPX8* display a lateral root density significantly higher than that of wild type, whereas in lines knock-out for *AtGPX2* and *AtGPX3*, this density is lower [209]. Remarkably, the expression of At*GPX4* and *AtGPX7* genes is induced by auxin, which has a recognized role in the development of lateral roots. This argues for the existence of interplays between hormones, ROS and GPXs in the control of root architecture. Taken collectively, the data gained in cereals and in the plant model Arabidopsis give strong credence for a key role of GPXs in the development of plant roots. Besides, the participation of GPXs in *A. thaliana* shoot development was highlighted in a mutant deficient in the expression of the two genes coding for plastidial GPXs, *AtGPX1* and *AtGPX7* [210]. Both leaf cell structure and chloroplast morphology are modified in the double mutant compared to wild-type plants, as well as its responses to environmental constraints. Another GPX seems to have a very crucial role. Indeed, in a study looking for genes required for female gametophyte development, an *A. thaliana* transposon insertion line in the *AtGPX5* gene has been found to be arrested at the embryo stage due to a defect in endosperm development [211]. Whether this role is related to ROS or hormonal signaling is not yet elucidated. Finally, it is worth mentioning that deletion of *AtGPX3* gene perturbs the response to ABA in guard cells and the expression of genes related to stress [212]. Moreover the *gpx3* mutant development is impaired in vitro in osmotic and oxidative stress conditions. Thus, AtGPX3 seems to be at the crossing between ABA-dependent stress responses and regulation of redox homeostasis.

*3.5.3. Roles of methionine sulfoxide reductases in plant development*

Methionine oxidation to methionine sulfoxide is reversed by A- and B-types MSRs, the regeneration of which is generally achieved by TRXs [183]. The Arabidopsis genome contains five *MSRA* and nine *MSRB* genes. With regard to MSRAs, an Arabidopsis line deficient for *AtMSRA2* displays reduced growth and slower development specifically in short-day conditions [213]. This isoform has been proposed to repair damaged proteins in the dark phase, thus preventing the loss of resources due to increased protein turnover. Two MSRB isoforms, AtMSRB1 and AtMSRB2, are present in *A. thaliana* chloroplasts. When cultivated in control conditions, Arabidopsis plants lacking plastidial AtMSRBs do not exhibit any phenotype whereas under high light or at low temperature, these plants display reduced growth, pale green leaves and lower seed yield compared to wild type [214]. Plastidial MSRBs have been proposed to protect the photosynthetic antennae from oxidative damage and thus to preserve growth and development in environmental stress conditions. Other Arabidopsis MSRB isoforms, AtMSRB7 and AtMSRB8, are preferentially expressed in roots [215]. Interestingly, Arabidopsis lines overexpressing these two isoforms show a noticeably reduced primary root length when grown in vitro in control conditions [215] unveiling a possible role of the control of Met redox status in root development. In pepper, the silencing of a gene encoding a CaMSRB2 isoform leads to strongly retarded growth and reduced development in addition to modified sensitivity to pathogen attack [216].

All these data reveal relatively unexpected roles of thiol peroxidases and MSRs in plant development even in the absence of environmental constraints. They give strong credence for a participation of these reductases in developmental processes probably as mediators of the cell redox homeostasis in relation with other actors such as hormones and ROS. Thus TRXs and their targets are likely to act in the mechanisms governing the phenotypic plasticity of plants in relation with the climate variations [217]. For example, those located in the plastidial compartment could act as sensors or participate in transduction pathways regulating nuclear gene expression. These pathways, known as retrograde signaling, establish relationships between the photosynthetic excitation pressure, which highly varies depending on environmental conditions, and the developmental processes thus enabling plant adaptation to changing conditions.

## 4. Conclusions

As shown in this review, GSH, TRXs and GRXs fulfill multiple and essential roles in the plant developmental cycle (Fig. 3). These roles have been demonstrated mainly on the basis of genetic approaches using mutants either defective in the synthesis of glutathione or deficient in the expression of one of the numerous plant *GRX* and *TRX* genes. Of note, cross-talks between thiol compounds and phytohormones like auxins, which are master regulators of plant cell differentiation, have been established. The participation of thiol-containing compounds in plant developmental processes is very probably tightly connected to other redox actors, such as reactive oxygen and nitrogen species, the contribution of which is central to the maintenance of an adequate cellular redox homeostasis. Compared to animal cells, examples showing the involvement of reactive species like $O_2^{\cdot-}$, $H_2O_2$ and NO in specific developmental stages are relatively limited in plants [73,74,197]. But as Cys residues in proteins are highly prone to react with most ROS and RNS [218,219], it is tempting to speculate that the participation of thiol-based mechanisms in the plant developmental program is partly linked to spatial or temporal variations in ROS and RNS concentrations. On the other hand, the role of NADPH, the electron donor to both TRX and GRX pathways, has also to be further investigated with regard to plant development. Indeed, tobacco plants modified for the expression of a mitochondrial calcium-dependent NADPH dehydrogenase display altered NADPH/$NADP^+$ ratio in stem, concomitantly to deregulated expression of flowering-associated genes and modified bolting transition [220]. This will be of great interest to decipher whether this NADPH function is fulfilled via TRXs/GRXs or other redox signaling transduction pathways. With regard to glutathione, a major actor with ascorbate in plant cell redox homeostasis, this tripeptide participates in the control of development at least through three functions: redox buffer, supply of reducing power to GRXs and post-translational modification of Cys residues in proteins via glutathionylation. Consistently, a large variety of defects has been observed in glutathione-deficient mutants, highlighting the crucial importance of this compound for plant development and stress response. Glutathione is thus likely to regulate directly or indirectly the redox status of many cysteine-containing proteins [5]. For example, cysteine glutathionylation might alter the activity or conformation of actors specifically involved in signaling transduction pathways, regulation of gene transcription or protein translation. Moreover, from the finding made in yeast that GSH is critical for the maturation of Fe–S clusters, prosthetic groups found in many essential proteins, we can assume that GSH essentiality in plant development relies on this point too.

Altogether, these findings strongly support the participation of GSH, GRXs and TRXs in the complex redox-based network governing plant development [221,222]. Most interestingly, it has been recently proposed that the plant cell could be considered as a set of compartments distinct in their capacities for ROS generation and antioxidant buffering [223]. This view is illustrated by the sequestration of GSSG in the vacuole, the highly reducing environment in the cytosol and the relatively oxidizing conditions reigning in the apoplast and cell wall [223]. With the development of new genetically-encoded probes able to perform dynamic and real-time measurements of the distribution and levels of glutathione [41], $H_2O_2$ [224] or auxin [225] in specific cell types or subcellular compartments, significant breakthroughs should be achieved in the future. Redox-sensitive actors are thus likely to play crucial functions in relaying signals between the cellular subcompartments and TRXs and GRXs are good candidates to act as sensors or transmitters in such transduction pathways via notably transient and reversible post-translational modifications of their thiol groups like disulfide bond formation, nitrosylation or glutathionylation. Nevertheless at the present time, the experimental evidence supporting this hypothesis are still poor except in the case of class III GRXs, which are able to interact with TGA transcription factors involved in the genesis of floral organs and to regulate their activity likely via redox-based

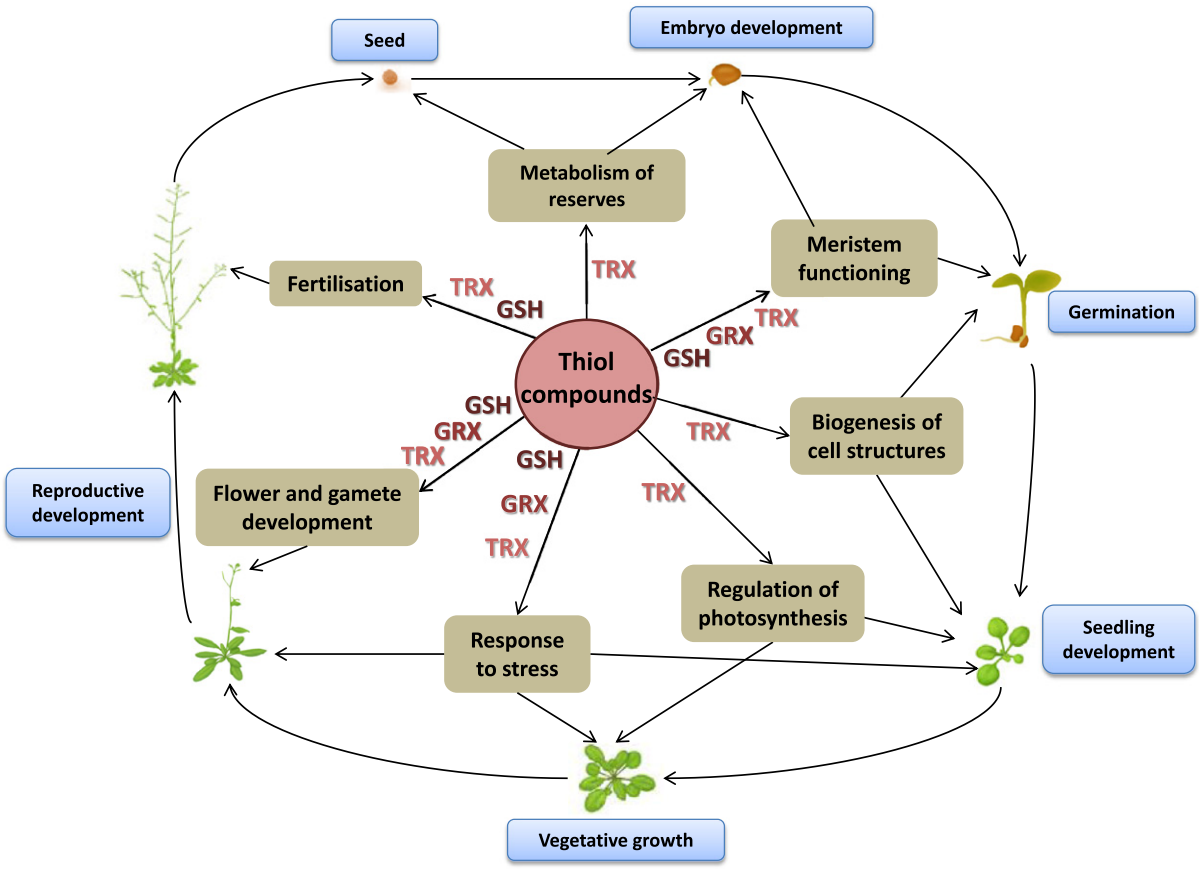


**Fig. 3.** Participation of GSH, GRXs and TRXs in the plant developmental cycle.

mechanisms [108,112]. Many TRX and GRX partners have been identified using biochemical strategies (thiol labeling, mutagenesis of active site residues and affinity chromatography) [120,226]. Among more than 400 putative partners of TRXs identified in land plants, very few at first sight appear as implicated in mechanisms related to development except some auxin-related proteins, RNA-binding proteins and transcriptional elongation factors. This is rather surprising since the activity of several types of transcriptional factors is likely controlled via the redox status of their cysteines as mentioned in the Chapter 3.2.2.1. The absence of nuclear factors among potential TRX partners could originate from the under-representation of nuclear proteins in the plant extracts used in the approaches set up for partner targeting. Applying these approaches on subcellular extracts or on extracts prepared from specific cell types, in combination with the ever more sensitive and sophisticated proteomic methods, will allow gaining insight into the roles of TRXs in the control of plant development.

Finally, it appears likely that disulfide reductases participate in plant development through much more unexpected roles, which remain to be discovered. Thus, a physiological function related to development has been unveiled for the well-known plastidial TRX-regulated protein CP12, which forms a complex with Calvin–Benson cycle enzymes. Indeed, *CP12* antisense tobacco plants exhibit very severe morphological changes, reduced growth rate and impaired fertility that are associated with modifications in carbon partitioning [227]. This report suggests the existence of complex transduction pathways linking carbon metabolism to developmental programs. The redox status of carbon assimilation enzymes might constitute an important signal for the proper progress of plant development phases which ultimately depend on energy availability.

## Transparency document

The Transparency document associated with this article can be found, in the online version.

## Acknowledgments

The work of N.R., J.C., J-P.R. and P.R. was supported by a grant from the French National Research Agency (ANR 2010 BLAN_1616). The UMR1136 is supported by a grant overseen by the French National Research Agency (ANR) as a part of the "Investissements d'Avenir" program (ANR-11-LABX-0002-01, Lab of Excellence ARBRE). JPR acknowledges support from the French National Research Agency (ANR-BLAN 12-BSV6-0011). DC acknowledges support from the "Région Provence-Alpes-Côte d'Azur" (PhD grant DEB 12-1538).

## References

[1] L.B. Poole, P.A. Karplus, A. Claiborne, Protein sulfenic acids in redox signaling, Annu. Rev. Pharmacol. Toxicol. 44 (2004) 325–347.
[2] L.M. Landino, Protein thiol modification by peroxynitrite anion and nitric oxide donors, Methods Enzymol. 440 (2008) 95–109.
[3] G. Noctor, C.H. Foyer, ASCORBATE AND GLUTATHIONE: keeping active oxygen under control, Annu. Rev. Plant Physiol. Plant Mol. Biol. 49 (1998) 249–279.
[4] A.J. Meyer, M.J. May, M. Fricker, Quantitative in vivo measurement of glutathione in Arabidopsis cells, Plant J. 27 (2001) 67–78.
[5] N. Rouhier, S.D. Lemaire, J.P. Jacquot, The role of glutathione in photosynthetic organisms: emerging functions for glutaredoxins and glutathionylation, Annu. Rev. Plant Biol. 59 (2008) 143–166.
[6] N. Rouhier, Plant glutaredoxins: pivotal players in redox biology and iron-sulphur centre assembly, New Phytol. 186 (2010) 365–372.

[7] N. Rouhier, E. Gelhaye, J.P. Jacquot, Plant glutaredoxins: still mysterious reducing systems, Cell. Mol. Life Sci. 61 (2004) 1266–1277.
[8] J. Couturier, J.P. Jacquot, N. Rouhier, Evolution and diversity of glutaredoxins in photosynthetic organisms, Cell. Mol. Life Sci. 66 (2009) 2539–2557.
[9] K. Chibani, G. Wingsle, J.P. Jacquot, E. Gelhaye, N. Rouhier, Comparative genomic study of the thioredoxin family in photosynthetic organisms with emphasis on *Populus trichocarpa*, Mol. Plant 2 (2009) 308–322.
[10] M. Zaffagnini, L. Michelet, V. Massot, P. Trost, S.D. Lemaire, Biochemical characterization of glutaredoxins from *Chlamydomonas reinhardtii* reveals the unique properties of a chloroplastic CGFS-type glutaredoxin, J. Biol. Chem. 283 (2008) 8868–8876.
[11] A. Holmgren, Thioredoxin, Annu. Rev. Biochem. 54 (1985) 237–271.
[12] S. Dai, C. Schwendtmayer, P. Schürmann, S. Ramaswamy, H. Eklund, Redox signaling in chloroplasts: cleavage of disulfides by an iron-sulfur cluster, Science 287 (2000) 655–658.
[13] E. Gelhaye, N. Rouhier, J. Gérard, Y. Jolivet, J. Gualberto, N. Navrot, P.I. Ohlsson, G. Wingsle, M. Hirasawa, D.B. Knaff, H. Wang, P. Dizengremel, Y. Meyer, J.P. Jacquot, A specific form of thioredoxin h occurs in plant mitochondria and regulates the alternative oxidase, Proc. Natl. Acad. Sci. U. S. A. 101 (2004) 14545–14550.
[14] C.S. Koh, N. Navrot, C. Didierjean, N. Rouhier, M. Hirasawa, D.B. Knaff, G. Wingsle, R. Samian, J.P. Jacquot, C. Corbier, E. Gelhaye, An atypical catalytic mechanism involving three cysteines of thioredoxin.J, Biol. Chem. 283 (2008) 23062–23072.
[15] L. Meng, J.H. Wong, L.J. Feldman, P.G. Lemaux, B.B. Buchanan, A membrane-associated thioredoxin required for plant growth moves from cell to cell, suggestive of a role in intercellular communication, Proc. Natl. Acad. Sci. U. S. A. 107 (2010) 3900–3905.
[16] K. Chibani, L. Tarrago, J.M. Gualberto, G. Wingsle, P. Rey, J.P. Jacquot, N. Rouhier, Atypical thioredoxins in poplar: the glutathione-dependent thioredoxin-like 2.1 supports the activity of target enzymes possessing a single redox active cysteine, Plant Physiol. 159 (2012) 592–605.
[17] R.A. Wolosiuk, B.B. Buchanan, Thioredoxin and glutathione regulate photosynthesis in chloroplasts, Nature 266 (1977) 565–567.
[18] K. Kobrehel, J.H. Wong, A. Balogh, F. Kiss, B.C. Yee, B.B. Buchanan, Specific reduction of wheat storage proteins by thioredoxin h, Plant Physiol. 99 (1992) 919–924.
[19] C. Vieira Dos Santos, P. Rey, Plant thioredoxins are key actors in oxidative stress response, Trends Plant Sci. 11 (2006) 329–334.
[20] S. Xing, M.G. Rosso, S. Zachgo, ROXY1, a member of the plant glutaredoxin family, is required for petal development in *Arabidopsis thaliana*, Development 132 (2005) 1555–1565.
[21] S. Xing, S. Zachgo, ROXY1 and ROXY2, two Arabidopsis glutaredoxin genes, are required for anther development, Plant J. 53 (2008) 790–801.
[22] N.H. Cheng, J.Z. Liu, X. Liu, Q. Wu, S.M. Thompson, J. Lin, J. Chang, S.A. Whitham, S. Park, J.D. Cohen, K.D. Hirschi, Arabidopsis monothiol glutaredoxin, AtGRXS17, is critical for temperature-dependent postembryonic growth and development via modulating auxin response, J. Biol. Chem. 286 (2011) 20398–20406.
[23] S. La Camera, F. L'Haridon, J. Astier, M. Zander, E. Abou-Mansour, G. Page, C. Thurow, D. Wendehenne, C. Gatz, J.P. Metraux, O. Lamotte, The glutaredoxin ATGRXS13 is required to facilitate *Botrytis cinerea* infection of *Arabidopsis thaliana* plants, Plant J. 68 (2011) 507–519.
[24] A. Wachter, S. Wolf, H. Steininger, J. Bogs, T. Rausch, Differential targeting of GSH1 and GSH2 is achieved by multiple transcription initiation: implications for the compartmentation of glutathione biosynthesis in the Brassicaceae, Plant J. 41 (2005) 15–30.
[25] M. Pasternak, B. Lim, M. Wirtz, R. Hell, C.S. Cobbett, A.J. Meyer, Restricting glutathione biosynthesis to the cytosol is sufficient for normal plant development, Plant J. 53 (2008) 999–1012.
[26] N.G. Cairns, M. Pasternak, A. Wachter, C.S. Cobbett, A.J. Meyer, Maturation of arabidopsis seeds is dependent on glutathione biosynthesis within the embryo, Plant Physiol. 141 (2006) 446–455.
[27] P. Frendo, J. Harrison, C. Norman, M.J. Hernández Jiménez, G. Van de Sype, A. Gilabert, A. Puppo, Glutathione and homoglutathione play a critical role in the nodulation process of *Medicago truncatula*, Mol. Plant Microbe Interact. 18 (2005) 254–259.
[28] S.C. Maughan, M. Pasternak, N. Cairns, G. Kiddle, T. Brach, R. Jarvis, F. Haas, J. Nieuwland, B. Lim, C. Müller, E. Salcedo-Sora, C. Kruse, M. Orsel, R. Hell, A.J. Miller, P. Bray, C.H. Foyer, J.A. Murray, A.J. Meyer, C.S. Cobbett, Plant homologs of the *Plasmodium falciparum* chloroquine-resistance transporter, PfCRT, are required for glutathione homeostasis and stress responses, Proc. Natl. Acad. Sci. U. S. A. 107 (2010) 2331–2336.
[29] T. Vernoux, R.C. Wilson, K.A. Seeley, J.P. Reichheld, S. Muroy, S. Brown, S.C. Maughan, C.S. Cobbett, M. Van Montagu, D. Inzé, M.J. May, Z.R. Sung, The ROOT MERISTEMLESS1/CADMIUM SENSITIVE2 gene defines a glutathione-dependent pathway involved in initiation and maintenance of cell division during postembryonic root development, Plant Cell 12 (2000) 97–110.
[30] R. Howden, C.R. Andersen, P.B. Goldsbrough, C.S. Cobbett, A cadmium-sensitive, glutathione-deficient mutant of *Arabidopsis thaliana*, Plant Physiol. 107 (1995) 1067–1073.
[31] L. Ball, G.P. Accotto, U. Bechtold, G. Creissen, D. Funck, A. Jimenez, B. Kular, N. Leyland, J. Mejia-Carranza, H. Reynolds, S. Karpinski, P.M. Mullineaux, Evidence for a direct link between glutathione biosynthesis and stress defense gene expression in Arabidopsis, Plant Cell 16 (2004) 2448–2462.
[32] V. Parisy, B. Poinssot, L. Owsianowski, A. Buchala, J. Glazebrook, F. Mauch, Identification of PAD2 as a gamma-glutamylcysteine synthetase highlights the importance of glutathione in disease resistance of Arabidopsis, Plant J. 49 (2007) 159–172.
[33] V. Shanmugam, M. Tsednee, K.C. Yeh, ZINC TOLERANCE INDUCED BY IRON 1 reveals the importance of glutathione in the cross-homeostasis between zinc and iron in *Arabidopsis thaliana*, Plant J. 69 (2012) 1006–1017.
[34] J.C. Cheng, K.A. Seeley, Z.R. Sung, RML1 and RML2, Arabidopsis genes required for cell proliferation at the root tip, Plant Physiol. 107 (1995) 365–376.
[35] J.P. Reichheld, M. Khafif, C. Riondet, M. Droux, G. Bonnard, Y. Meyer, Inactivation of thioredoxin reductases reveals a complex interplay between thioredoxin and glutathione pathways in Arabidopsis development, Plant Cell 19 (2007) 1851–1865.
[36] C.S. Cobbett, M.J. May, R. Howden, B. Rolls, The glutathione-deficient, cadmium-sensitive mutant, cad2-1, of *Arabidopsis thaliana* is deficient in gamma-glutamylcysteine synthetase, Plant J. 16 (1998) 73–78.
[37] B. Zechmann, B.E. Koffler, S.D. Russell, Glutathione synthesis is essential for pollen germination *in vitro*, BMC Plant Biol. 11 (2011) 1–11.
[38] L. Marty, W. Siala, M. Schwarzländer, M.D. Fricker, M. Wirtz, L.J. Sweetlove, Y. Meyer, A.J. Meyer, J.P. Reichheld, R. Hell, The NADPH-dependent thioredoxin system constitutes a functional backup for cytosolic glutathione reductase in Arabidopsis, Proc. Natl. Acad. Sci. U. S. A. 106 (2009) 9109–9114.
[39] T. Bashandy, J. Guilleminot, T. Vernoux, D. Caparros-Ruiz, K. Ljung, Y. Meyer, J.P. Reichheld, Interplay between the NADP-linked thioredoxin and glutathione systems in Arabidopsis auxin signalling, Plant Cell 22 (2010) 376–391.
[40] M. Hothorn, A. Wachter, R. Gromes, T. Stuwe, T. Rausch, K. Scheffzek, Structural basis for the redox control of plant glutamate cysteine ligase, J. Biol. Chem. 281 (2006) 27557–27565.
[41] A.J. Meyer, T. Brach, L. Marty, S. Kreye, N. Rouhier, J.P. Jacquot, R. Hell, Redox-sensitive GFP in *Arabidopsis thaliana* is a quantitative biosensor for the redox potential of the cellular glutathione redox buffer, Plant J. 52 (2007) 973–986.
[42] I. Aller, N. Rouhier, A.J. Meyer, Development of roGFP2-derived redox probes for measurement of the glutathione redox potential in the cytosol of severely glutathione-deficient rml1 seedlings, Front. Plant Sci. 4 (2013) 506.
[43] X. Yu, T. Pasternak, M. Eiblmeier, F. Ditengou, P. Kochersperger, J. Sun, H. Wang, H. Rennenberg, W. Teale, I. Paponov, W. Zhou, C. Li, X. Li, K. Palme, Plastid-localized glutathione reductase2-regulated glutathione redox status is essential for Arabidopsis root apical meristem maintenance, Plant Cell 25 (2013) 4451–4468.
[44] I. Tzafrir, R. Pena-Muralla, A. Dickerman, M. Berg, R. Rogers, S. Hutchens, T.C. Sweeney, J. McElver, G. Aux, D. Patton, D. Meinke, Identification of genes required for embryo development in Arabidopsis, Plant Physiol. 135 (2004) 1206–1220.
[45] T.A. Schaedler, J.D. Thornton, I. Kruse, M. Schwarzländer, A.J. Meyer, H.W. van Veen, J. Balk, A conserved mitochondrial ATP-binding cassette transporter exports glutathione polysulfide for cytosolic metal cofactor assembly, J. Biol. Chem. 289 (2014) 23264–23674.
[46] J.P. Shaw, I.N. Chou, Elevation of intracellular glutathione content associated with mitogenic stimulation of quiescent fibroblasts, J. Cell. Physiol. 129 (1986) 193–198.
[47] N. Kerk, L. Feldman, A biochemical model for the initiation and maintenance of the quiescent center: implications for organization of root meristems, Development 121 (1995) 2825–2833.
[48] R. Sánchez-Fernández, M. Fricker, L.B. Corben, N.S. White, N. Sheard, C.J. Leaver, M. Van Montagu, D. Inzé, M.J. May, Cell proliferation and hair tip growth in the Arabidopsis root are under mechanistically different forms of redox control, Proc. Natl. Acad. Sci. U. S. A. 94 (1997) 2745–2750.
[49] G. Potters, N. Horemans, S. Bellone, R.J. Caubergs, P. Trost, Y. Guisez, H. Asard, Dehydroascorbate influences the plant cell cycle through a glutathione-independent reduction mechanism, Plant Physiol. 134 (2004) 1479–1487.
[50] I. Kranner, S. Birtic, K.M. Anderson, H.W. Pritchard, Glutathione half-cell reduction potential: a universal stress marker and modulator of programmed cell death, Free Radic. Biol. Med. 40 (2006) 2155–2165.
[51] K. Jiang, Y.L. Meng, L.J. Feldman, Quiescent center formation in maize roots is associated with an auxin-regulated oxidizing environment, Development 130 (2003) 1429–1438.
[52] J. Markovic, C. Borrás, A. Ortega, J. Sastre, J. Viña, F.V. Pallardó, Glutathione is recruited into the nucleus in early phases of cell proliferation, J. Biol. Chem. 282 (2007) 20416–20424.
[53] P. Diaz Vivancos, Y. Dong, K. Ziegler, J. Markovic, F.V. Pallardó, T.K. Pellny, P.J. Verrier, C.H. Foyer, Recruitment of glutathione into the nucleus during cell proliferation adjusts whole-cell redox homeostasis in *Arabidopsis thaliana* and lowers the oxidative defence shield, Plant J. 64 (2010) 825–838.
[54] J. Markovic, N.J. Mora, A.M. Broseta, A. Gimeno, N. de-la-Concepción, J. Viña, F.V. Pallardó, The depletion of nuclear glutathione impairs cell proliferation in 3t3 fibroblasts, PLoS One 4 (2009) e6413.
[55] P. Diaz Vivancos, T. Wolff, J. Markovic, F.V. Pallardó, C.H. Foyer, A nuclear glutathione cycle within the cell cycle, Biochem. J. 431 (2010) 169–178.
[56] T. Pasternak, H. Asard, G. Potters, M.A. Jansen, The thiol compounds glutathione and homoglutathione differentially affect cell development in alfalfa (*Medicago sativa* L.), Plant Physiol. Biochem. 74 (2014) 16–23.
[57] A. Koornneef, A. Leon-Reyes, T. Ritsema, A. Verhage, F.C. Den Otter, L.C. Van Loon, C.M.J. Pieterse, Kinetics of salicylate-mediated suppression of jasmonate signaling reveal a role for redox modulation, Plant Physiol. 147 (2008) 1358–1368.
[58] A. Mhamdi, J. Hager, S. Chaouch, G. Queval, Y. Han, L. Taconnat, P. Saindrenan, H. Gouia, E. Issakidis-Bourguet, J.P. Renou, G. Noctor, Arabidopsis glutathione reductase1 plays a crucial role in leaf responses to intracellular hydrogen peroxide and in ensuring appropriate gene expression through both salicylic acid and jasmonic acid signaling pathways, Plant Physiol. 153 (2010) 1144–1160.
[59] Y. Han, A. Mhamdi, S. Chaouch, G. Noctor, Regulation of basal and oxidative stress-triggered jasmonic acid-related gene expression by glutathione, Plant Cell Environ. 36 (2013) 1135–1146.
[60] I. Ndamukong, A.A. Abdallat, C. Thurow, B. Fode, M. Zander, R. Weigel, C. Gatz, SA-inducible Arabidopsis glutaredoxin interacts with TGA factors and suppresses JA-responsive PDF1.2 transcription, Plant J. 50 (2007) 128–139.

[61] M. Dron, S.D. Clouse, R.A. Dixon, M.A. Lawton, C.J. Lamb, Glutathione and fungal elicitor regulation of a plant defense gene promoter in electroporated protoplasts, Proc. Natl. Acad. Sci. U. S. A. 85 (1988) 6738–6742.
[62] R. Edwards, J.W. Blount, R.A. Dixon, Glutathione and elicitation of the phytoalexin response in legume cell cultures, Planta 184 (1991) 403–409.
[63] Z. Mou, W. Fan, X. Dong, Inducers of plant systemic acquired resistance regulate NPR1 function through redox changes, Cell 113 (2003) 935–944.
[64] C. Després, C. Chubak, A. Rochon, R. Clark, T. Bethune, D. Desveaux, P.R. Fobert, The Arabidopsis NPR1 disease resistance protein is a novel cofactor that confers redox regulation of DNA binding activity to the basic domain/leucine zipper transcription factor TGA1, Plant Cell 15 (2003) 2181–2191.
[65] A. Rochon, P. Boyle, T. Wignes, P.R. Fobert, C. Després, The coactivator function of Arabidopsis NPR1 requires the core of its BTB/POZ domain and the oxidation of C-terminal oxidases, Plant Cell 18 (2006) 3670–3685.
[66] Y. Tada, S.H. Spoel, K. Pajerowska-Mukhtar, Z. Mou, J. Song, X. Dong, Plant immunity requires conformational changes of NPR1 via S-nitrosylation and thioredoxins, Science 321 (2008) 952–956.
[67] S. Kneeshaw, S. Gelineau, Y. Tada, G.J. Loake, S.H. Spoel, Selective protein denitrosylation activity of thioredoxin-h5 modulates plant immunity, Mol. Cell 56 (2014) 153–162.
[68] G. Noctor, G. Queval, A. Mhamdi, S. Chaouch, C.H. Foyer, Glutathione, Arabidopsis Book 9 (2011) e0142.
[69] E. Okuma, M.S. Jahan, S. Munemasa, M.A. Hossain, D. Muroyama, M.M. Islam, K. Ogawa, M. Watanabe-Sugimoto, Y. Nakamura, Y. Shimoishi, I.C. Mori, Y. Murata, Negative regulation of abscisic acid-induced stomatal closure by glutathione in Arabidopsis, J. Plant Physiol. 168 (2011) 2048–2055.
[70] K. Jiang, T. Zhu, Z. Diao, H. Huang, L.J. Feldman, The maize root stem cell niche: a partnership between two sister cell populations, Planta 231 (2010) 411–424.
[71] A. Koprivova, S.T. Mugford, S. Kopriva, Arabidopsis root growth dependence on glutathione is linked to auxin transport, Plant Cell Rep. 29 (2010) 1157–1167.
[72] D. Schnaubelt, G. Queval, Y. Dong, P. Diaz-Vivancos, M.E. Makgopa, G. Howell, A. DE Simone, J. Bai, M.A. Hannah, C.H. Foyer, Low glutathione regulates gene expression and the redox potentials of the nucleus and cytosol in *Arabidopsis thaliana*, Plant Cell Environ. (2013)http://dx.doi.org/10.1111/pce.12252.
[73] M.C. Terrile, R. París, L.I. Calderón-Villalobos, M.J. Iglesias, L. Lamattina, M. Estelle, C.A. Casalongué, Nitric oxide influences auxin signaling through S-nitrosylation of the Arabidopsis TRANSPORT INHIBITOR RESPONSE 1 auxin receptor, Plant J. 70 (2012) 492–500.
[74] M. Fernández-Marcos, L. Sanz, D.R. Lewis, G.K. Muday, O. Lorenzo, Nitric oxide causes root apical meristem defects and growth inhibition while reducing PIN-FORMED 1 (PIN1)-dependent acropetal auxin transport, Proc. Natl. Acad. Sci. U. S. A. 108 (2011) 18506–18511.
[75] T. Blomster, J. Salojärvi, N. Sipari, M. Brosché, R. Ahlfors, M. Keinänen, K. Overmyer, J. Kangasjärvi, Apoplastic reactive oxygen species transiently decrease auxin signaling and cause stress-induced morphogenic response in Arabidopsis, Plant Physiol. 157 (2011) 1866–1883.
[76] X. Gao, H.M. Yuan, Y.Q. Hu, J. Li, Y.T. Lu, Mutation of Arabidopsis CATALASE2 results in hyponastic leaves by changes of auxin levels, Plant Cell Environ. 37 (2014) 175–188.
[77] P. Kerchev, P. Mühlenbock, J. Denecker, K. Morreel, F. Hoeberichts, K. Van Der Kelen, M. Vandorpe, L. Nguyen, D. Audenaert, F. Van Breusegem, Activation of auxin signalling counteracts photorespiratory $H_2O_2$-dependent cell death, Plant Cell Environ. (2014)http://dx.doi.org/10.1111/pce.12250.
[78] V.B. Tognetti, O. Van Aken, K. Morreel, K. Vandenbroucke, B. van de Cotte, I. De Clercq, S. Chiwocha, R. Fenske, E. Prinsen, W. Boerjan, B. Genty, K.A. Stubbs, D. Inzé, F. Van Breusegem, Perturbation of indole-3-butyric acid homeostasis by the UDP-glucosyltransferase UGT74E2 modulates Arabidopsis architecture and water stress tolerance, Plant Cell 22 (2010) 2660–2679.
[79] B. Marquez-Garcia, M. Njo, T. Beeckman, S. Goormachtig, C.H. Foyer, A new role for glutathione in the regulation of root architecture linked to strigolactones, Plant Cell Environ. 37 (2014) 488–498.
[80] K. Sipos, H. Lange, Z. Fekete, P. Ullmann, R. Lill, G. Kispal, Maturation of cytosolic iron-sulfur proteins requires glutathione, J. Biol. Chem. 277 (2002) 26944–26949.
[81] C. Kumar, A. Igbaria, B. D'Autreaux, A.G. Planson, C. Junot, E. Godat, A.K. Bachhawat, A. Delaunay-Moisan, M.B. Toledano, Glutathione revisited: a vital function in iron metabolism and ancillary role in thiol-redox control, EMBO J. 30 (2011) 2044–2056.
[82] J. Couturier, B. Touraine, J.F. Briat, F. Gaymard, N. Rouhier, The iron-sulfur cluster assembly machineries in plants: current knowledge and open questions, Front. Plant Sci. 4 (2013) 259.
[83] N. Rouhier, H. Unno, S. Bandyopadhyay, L. Masip, S.K. Kim, M. Hirasawa, J.M. Gualberto, V. Lattard, M. Kusunoki, D.B. Knaff, G. Georgiou, T. Hase, M.K. Johnson, J.P. Jacquot, Functional, structural, and spectroscopic characterization of a glutathione-ligated [2Fe-2S] cluster in poplar glutaredoxin C1, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 7379–7384.
[84] R. Lill, Function and biogenesis of iron-sulphur proteins, Nature 460 (2009) 831–838.
[85] J.H. Chen, H.W. Jiang, E.J. Hsieh, H.Y. Chen, C.T. Chien, H.L. Hsieh, T.P. Lin, Drought and salt stress tolerance of an Arabidopsis glutathione S-transferase U17 knockout mutant are attributed to the combined effect of glutathione and abscisic acid, Plant Physiol. 158 (2012) 340–351.
[86] H.W. Jiang, M.J. Liu, I.C. Chen, C.H. Huang, L.Y. Chao, H.L. Hsieh, A glutathione S-transferase regulated by light and hormones participates in the modulation of Arabidopsis seedling development, Plant Physiol. 154 (2010) 1646–1658.
[87] Z. Chen, D.R. Gallie, Dehydroascorbate reductase affects leaf growth, development, and function, Plant Physiol. 142 (2006) 775–787.
[88] X.H. Gao, M. Bedhomme, D. Veyel, M. Zaffagnini, S.D. Lemaire, Methods for analysis of protein glutathionylation and their application to photosynthetic organisms, Mol. Plant 2 (2008) 218–235.
[89] Y. Meyer, W. Siala, T. Bashandy, C. Riondet, F. Vignols, J.P. Reichheld, Glutaredoxins and thioredoxins in plants, Biochim. Biophys. Acta 1783 (2008) 589–600.
[90] M. Ziemann, M. Bhave, S. Zachgo, Origin and diversification of land plant CC-type glutaredoxins, Genome Biol. Evol. 31 (2009) 265–267.
[91] P. Wang, J. Liu, B. Liu, D. Feng, Q. Da, P. Wang, S. Shu, J. Su, Y. Zhang, J. Wang, H.B. Wang, Evidence for a role of chloroplastic m-type thioredoxins in the biogenesis of photosystem II in Arabidopsis, Plant Physiol. 163 (2013) 1710–1728.
[92] Z. Wang, S. Xing, R.P. Birkenbihl, S. Zachgo, Conserved functions of Arabidopsis and rice CC-type glutaredoxins in flower development and pathogen response, Mol. Plant 2 (2009) 323–335.
[93] J.P. Reichheld, D. Mestres-Ortega, C. Laloi, Y. Meyer, The multigenic family of thioredoxin *h* in *Arabidopsis thaliana*: specific expression and stress response, Plant Physiol. Biochem. 40 (2002) 685–690.
[94] C. Belin, T. Bashandy, J. Cela, V. Delorme-Hinoux, C. Riondet, J.P. Reichheld, A comprehensive study of thiol reduction gene expression under stress conditions in *Arabidopsis thaliana*, Plant Cell Environ. (2015)http://dx.doi.org/10.1111/pce.12276.
[95] J.D. Becker, L.C. Boavida, J. Carneiro, M. Haury, J.A. Feijó, Transcriptional profiling of Arabidopsis tissues reveals the unique characteristics of the pollen transcriptome, Plant Physiol. 133 (2003) 713–725.
[96] J.Y. Lee, D.H. Lee, Use of serial analysis of gene expression technology to reveal changes in gene expression in Arabidopsis pollen undergoing cold stress, Plant Physiol. 132 (2003) 517–529.
[97] J.A. Traverso, F. Vignols, R. Cazalis, A. Pulido, M. Sahrawy, F.J. Cejudo, Y. Meyer, A. Chueca, PsTRXh1 and PsTRXh2 are both pea h-type thioredoxins with antagonistic behavior in redox imbalances, Plant Physiol. 143 (2007) 300–311.
[98] F. Montrichard, M. Renard, F. Alkhalfioui, F.D. Duval, D. Macherel, Identification and differential expression of two thioredoxin h isoforms in germinating seeds from pea, Plant Physiol. 132 (2003) 1707–1715.
[99] M. Renard, F. Alkhalfioui, C. Schmitt-Keichinger, C. Ritzenthaler, F. Montrichard, Identification and characterization of thioredoxin h isoforms differentially expressed in germinating seeds of the model legume *Medicago truncatula*, Plant Physiol. 155 (2011) 1113–1126.
[100] R. Cazalis, P. Pulido, T. Aussenac, J.M. Pérez-Ruiz, F.J. Cejudo, Cloning and characterization of three thioredoxin h isoforms from wheat showing differential expression in seeds, J. Exp. Bot. 57 (2006) 2165–2172.
[101] M.Y. Lee, K.H. Shin, Y.K. Kim, J.Y. Suh, Y.Y. Gu, M.R. Kim, Y.S. Hur, O. Son, J.S. Kim, E. Song, M.S. Lee, K.H. Nam, K.H. Hwang, M.K. Sung, H.J. Kim, J.Y. Chun, M. Park, T.I. Ahn, C.B. Hong, S.H. Lee, H.J. Park, J.S. Park, D.P. Verma, C.I. Cheon, Induction of thioredoxin is required for nodule development to reduce reactive oxygen species levels in soybean roots, Plant Physiol. 139 (2006) 1881–1889.
[102] F. Alkhalfioui, M. Renard, P. Frendo, C. Keichinger, Y. Meyer, E. Gelhaye, M. Hirasawa, D.B. Knaff, C. Ritzenthaler, F. Montrichard, A novel type of thioredoxin dedicated to symbiosis in legumes, Plant Physiol. 148 (2008) 424–435.
[103] J.A. Traverso, F. Vignols, R. Cazalis, A.J. Serrato, P. Pulido, M. Sahrawy, Y. Meyer, F.J. Cejudo, A. Chueca, Immunocytochemical localization of *Pisum sativum* TRXs f and m in non-photosynthetic tissues, J. Exp. Bot. 59 (2008) 1267–1277.
[104] C. Riondet, J.P. Desouris, J.G. Montoyan, Y. Chartier, Y. Meyer, J.P. Reichheld, A dicotyledon-specific glutaredoxin GRXC1 family with dimer-dependent redox regulation is functionally redundant with GRXC2, Plant Cell Environ. 35 (2012) 360–373.
[105] J. Couturier, J.P. Jacquot, N. Rouhier, Toward a refined classification of class I dithiol glutaredoxins from poplar: biochemical basis for the definition of two subclasses, Front. Plant Sci. 4 (2013) 518.
[106] J. Knuesting, C. Riondet, C. Maria, I. Kruse, N. Bécuwe, N. König, C. Berndt, S. Tourrette, J. Guilleminot-Montoya, E. Herrero, F. Gaymard, J. Balk, G. Belli, R. Scheibe, J.P. Reichheld, N. Rouhier, P. Rey, Arabidopsis glutaredoxin S17 and its partner NF-YC11/NC2α contribute to maintenance of the shoot apical meristem under long-day photoperiod, Plant Physiol. (2015) pp.00049. http://dx.doi.org/10.1104/pp.15.00049 (First Published on February 19, 2015).
[107] X. Hou, W.W. Hu, L. Shen, L.Y. Lee, Z. Tao, J.H. Han, H. Yu, Global identification of DELLA target genes during Arabidopsis flower development, Plant Physiol. 147 (2008) 1126–1142.
[108] L. Hong, D. Tang, K. Zhu, K. Wang, M. Li, Z. Cheng, Somatic and reproductive cell development in rice anther is regulated by a putative glutaredoxin, Plant Cell 24 (2012) 577–588.
[109] R. Chaubal, J.R. Anderson, M.R. Trimnell, T.W. Fox, M.C. Albertsen, P. Bedinger, The transformation of anthers in the msca1 mutant of maize, Planta 216 (2003) 778–788.
[110] T. Kelliher, V. Walbot, Hypoxia triggers meiotic fate acquisition in maize, Science 337 (2012) 345–348.
[111] L. Timofejeva, D.S. Skibbe, S. Lee, I. Golubovskaya, R. Wang, L. Harper, V. Walbot, W.Z. Cande, Cytological characterization and allelism testing of anther developmental mutants identified in a screen of maize male sterile lines, G3 (Bethesda) 3 (2013) 231–249.
[112] S. Li, A. Lauri, M. Ziemann, A. Busch, M. Bhave, S. Zachgo, Nuclear activity of ROXY1, a glutaredoxin interacting with TGA factors, is required for petal development in *Arabidopsis thaliana*, Plant Cell 21 (2009) 429–441.
[113] C.F. Chuang, M.P. Running, R.W. Williams, E.M. Meyerowitz, The PERIANTHIA gene encodes a bZIP protein involved in the determination of floral organ number in *Arabidopsis thaliana*, Genes Dev. 13 (1999) 334–344.

[114] J. Couturier, C. Didierjean, J.P. Jacquot, N. Rouhier, Engineered mutated glutaredoxins mimicking peculiar plant class III glutaredoxins bind iron–sulfur centers and possess reductase activity, Biochem. Biophys. Res. Commun. 403 (2010) 435–441.
[115] J. Murmu, M.J. Bush, C. DeLong, S. Li, M. Xu, M. Khan, C. Malcolmson, P.R. Fobert, S. Zachgo, S.R. Hepworth, Arabidopsis basic leucine-zipper transcription factors TGA9 and TGA10 interact with floral glutaredoxins ROXY1 and ROXY2 and are redundantly required for anther development, Plant Physiol. 154 (2010) 1492–1504.
[116] S. Li, N. Gutsche, S. Zachgo, The ROXY1 C-terminal L**LL motif is essential for the interaction with TGA transcription factors, Plant Physiol. 157 (2011) 2056–2068.
[117] M. Zander, S. Chen, J. Imkampe, C. Thurow, C. Gatz, Repression of the *Arabidopsis thaliana* jasmonic acid/ethylene-induced defense pathway by TGA-interacting glutaredoxins depends on their C-terminal ALWL motif, Mol. Plant 5 (2012) 831–840.
[118] R. Garg, S. Jhanwar, A.K. Tyagi, M. Jain, Genome-wide survey and expression analysis suggest diverse roles of glutaredoxin gene family members during development and response to various stimuli in rice, DNA Res. 17 (2010) 353–367.
[119] D. Laporte, E. Olate, P. Salinas, M. Salazar, X. Jordana, L. Holuigue, Glutaredoxin GRXS13 plays a key role in protection against photooxidative stress in Arabidopsis, J. Exp. Bot. 63 (2012) 503–515.
[120] F. Montrichard, F. Alkhalfioui, H. Yano, W.H. Vensel, W.J. Hurkman, B.B. Buchanan, Thioredoxin targets in plants: the first 30 years, J. Proteomics 72 (2009) 452–474.
[121] K.J. Dietz, Redox regulation of transcription factors in plant stress acclimation and development, Antioxid. Redox Signal. 21 (2014) 1356–1372.
[122] G.F. Heine, J.M. Hernandez, E. Grotewold, Two cysteines in plant R2R3 MYB domains participate in REDOX-dependent DNA binding, J. Biol. Chem. 279 (2004) 37878–37885.
[123] J. Shaikhali, L. Norén, Barajas-López J. de Dios, V. Srivastava, J. König, U.H. Sauer, G. Wingsle, K.J. Dietz, Å. Strand, Redox-mediated mechanisms regulate DNA binding activity of the G-group of basic region leucine zipper (bZIP) transcription factors in Arabidopsis, J. Biol. Chem. 287 (2012) 27510–27525.
[124] L. Turchi, M. Carabelli, V. Ruzza, M. Possenti, M. Sassi, A. Peñalosa, G. Sessa, S. Salvi, V. Forte, G. Morelli, I. Ruberti, Arabidopsis HD-Zip II transcription factors control apical embryo development and meristem function, Development 140 (2013) 2118–2129.
[125] J.R. McConnell, J. Emery, Y. Eshed, N. Bao, J. Bowman, M.K. Barton, Role of PHABULOSA and PHAVOLUTA in determining radial patterning in shoots, Nature 411 (2001) 709–713.
[126] R.N. Comelli, D.H. Gonzalez, Conserved homeodomain cysteines confer redox sensitivity and influence the DNA binding properties of plant class III HD-Zip proteins, Arch. Biochem. Biophys. 467 (2007) 41–47.
[127] I.L. Viola, L.N. Güttlein, D.H. Gonzalez, Redox modulation of plant developmental regulators from the class I TCP transcription factor family, Plant Physiol. 162 (2013) 1434–1447.
[128] Y. Benitez-Alfonso, M. Cilia, A. San Roman, C. Thomas, A. Maule, S. Hearn, D. Jackson, Control of Arabidopsis meristem development by thioredoxin-dependent regulation of intercellular transport, Proc. Natl. Acad. Sci. U. S. A. 106 (2009) 3615–3620.
[129] C.J. Zhang, B.C. Zhao, W.N. Ge, Y.F. Zhang, Y. Song, D.Y. Sun, Y. Guo, An apoplastic h-type thioredoxin is involved in the stress response through regulation of the apoplastic reactive oxygen species in rice, Plant Physiol. 157 (2011) 1884–1899.
[130] W. Azri, N. Brunel, J. Franchel, I. Ben Rejeb, J.P. Jacquot, J.L. Julien, S. Herbette, P. Roeckel-Drevet, Putative involvement of Thioredoxin h in early response to gravitropic stimulation of poplar stems, J. Plant Physiol. 170 (2013) 707–711.
[131] J.A. Traverso, A. Pulido, M.I. Rodríguez-García, J.D. Alché, Thiol-based redox regulation in sexual plant reproduction: new insights and perspectives, Front. Plant Sci. 4 (2013) 465.
[132] Y. Qin, A.R. Leydon, A. Manziello, R. Pandey, D. Mount, S. Denic, B. Vasic, M.A. Johnson, R. Palanivelu, Penetration of the stigma and style elicits a novel transcriptome in pollen tubes, pointing to genes critical for growth in a pistil, PLoS Genet. 8 (2009) e1000621.
[133] C. Marchal, V. Delorme-Hinoux, L. Bariat, W. Siala, C. Belin, J. Saez-Vasquez, C. Riondet, J.P. Reichheld, NTR/NRX define a new thioredoxin system in the nucleus of *Arabidopsis thaliana* cells, Mol. Plant 7 (2014) 30–44.
[134] M.S. Bower, D.D. Matias, E. Fernandes-Carvalho, M. Mazzurco, T. Gu, S.J. Rothstein, D.R. Goring, Two members of the thioredoxin-h family interact with the kinase domain of a Brassica S locus receptor kinase, Plant Cell 8 (1996) 1641–1650.
[135] D. Cabrillac, J.M. Cock, C. Dumas, T. Gaude, The S-locus receptor kinase is inhibited by thioredoxins and activated by pollen coat proteins, Nature 410 (2001) 220–223.
[136] R. Ivanov, T. Gaude, Endocytosis and endosomal regulation of the S-receptor kinase during the self-incompatibility response in Brassica oleracea, Plant Cell 21 (2009) 2107–2117.
[137] Y.Z. Haffani, T. Gaude, J.M. Cock, D.R. Goring, Antisense suppression of thioredoxin h mRNA in *Brassica napus* cv. Westar response, Plant Mol. Biol. 55 (2004) 619–630.
[138] M. Yamamoto, J.B. Nasrallah, In planta assessment of the role of thioredoxin h proteins in the regulation of S-locus receptor kinase signaling in transgenic Arabidopsis, Plant Physiol. 163 (2009) 1387–1395.
[139] J.A. Juárez-Díaz, B. McClure, S. Vázquez-Santana, A. Guevara-García, P. León-Mejía, J. Márquez-Guzmán, F. Cruz-García, A novel thioredoxin h is secreted in *Nicotiana alata* and reduces S-RNase *in vitro*, J. Biol. Chem. 281 (2006) 3418–3424.
[140] X. Li, J. Nield, D. Hayman, P. Langridge, Cloning a putative self-incompatibility gene from the pollen of the grass *Phalaris coerulescens*, Plant Cell 6 (1994) 1923–1932.
[141] X. Li, J. Nield, D. Hayman, P. Langridge, Thioredoxin activity in the C terminus of Phalaris S protein, Plant J. 8 (1995) 133–138.
[142] X. Li, J. Nield, D. Hayman, P. Langridge, A self-fertile mutant of Phalaris produces an S protein with reduced thioredoxin activity, Plant J. 10 (1996) 505–513.
[143] C. Lindermayr, S. Sell, B. Müller, D. Leister, J. Durner, Redox regulation of the NPR1-TGA1 system of *Arabidopsis thaliana* by nitric oxide, Plant Cell 22 (2010) 2894–2907.
[144] P. Schürmann, B.B. Buchanan, The ferredoxin/thioredoxin system of oxygenic photosynthesis, Antioxid. Redox Signal. 10 (2008) 1235–1274.
[145] E. Keryer, V. Collin, D. Lavergne, S. Lemaire, E. Issakidis-Bourguet, Characterization of Arabidopsis mutants for the variable subunit of ferredoxin:thioredoxin reductase, Photosynth. Res. 79 (2004) 265–274.
[146] P. Wang, J. Liu, B. Liu, Q. Da, D. Feng, J. Su, Y. Zhang, J. Wang, H.B. Wang, Ferredoxin: thioredoxin reductase Is required for proper chloroplast development and Is involved in the regulation of plastid gene expression in *Arabidopsis thaliana*, Mol. Plant (2014)http://dx.doi.org/10.1093/mp/ssu069.
[147] B. Arsova, U. Hoja, M. Wimmelbacher, E. Greiner, S. Ustün, M. Melzer, K. Petersen, W. Lein, F. Börnke, Plastidial thioredoxin z interacts with two fructokinase-like proteins in a thiol-dependent manner: evidence for an essential role in chloroplast development in Arabidopsis and *Nicotiana benthamiana*, Plant Cell 22 (2010) 1498–1515.
[148] K. Chibani, L. Tarrago, P. Schürmann, J.P. Jacquot, N. Rouhier, Biochemical properties of poplar thioredoxin z, FEBS Lett. 585 (2011) 1077–1081.
[149] A.S. Bohrer, V. Massot, G. Innocenti, J.P. Reichheld, E. Issakidis-Bourguet, H. Vanacker, New insights into the reduction systems of plastidial thioredoxins point out the unique properties of thioredoxin z from Arabidopsis, J. Exp. Bot. 63 (2012) 6315–6323.
[150] M. Wimmelbacher, F. Börnke, Redox activity of thioredoxin z and fructokinase-like protein 1 is dispensable for autotrophic growth of *Arabidopsis thaliana*, J. Exp. Bot. 65 (2014) 2405–2413.
[151] J. Qiao, C. Ma, M. Wimmelbacher, F. Bornke, M. Luo, Two novel proteins, MRL7 and its paralog MRL7-L, have essential but functionally distinct roles in chloroplast development and are involved in plastid gene expression regulation in Arabidopsis, Plant Cell Physiol. 52 (2011) 1017–1030.
[152] Q.B. Yua, Q. Ma, M.M. Kong, T.T. Zhao, X.L. Zhang, Q. Zhou, C. Huang, K. Chong, Z.N. Yang, AtECB1/MRL7, a thioredoxin-like fold protein with disulfide reductase activity, regulates chloroplast gene expression and chloroplast biogenesis in *Arabidopsis thaliana*, Mol. Plant 7 (2014) 206–217.
[153] Q.B. Yu, Y. Lu, Q. Ma, T.T. Zhao, C. Huang, H.F. Zhao, X.L. Zhang, R.H. Lv, Z.N. Yang, TAC7, an essential component of the plastid transcriptionally active chromosome complex, interacts with FLN1, TAC10, TAC12 and TAC14 to regulate chloroplast gene expression in Arabidopsis thaliana, Physiol. Plant. 148 (2013) 408–421.
[154] K. Motohashi, T. Hisabori, CcdA is a thylakoid membrane protein required for the transfer of reducing equivalents from stroma to thylakoid lumen in the higher plant chloroplast, Antioxid. Redox Signal. 13 (2010) 1169–1176.
[155] K. Lennartz, H. Plücken, A. Seidler, P. Westhoff, N. Bechtold, K. Meierhoff, HCF164 encodes a thioredoxin-like protein involved in the biogenesis of the cytochrome b(6)f complex in Arabidopsis, Plant Cell 13 (2001) 2539–2551.
[156] M.L. Page, P.P. Hamel, S.T. Gabilly, H. Zegzouti, J.V. Perea, J.M. Alonso, J.R. Ecker, S.M. Theg, S.K. Christensen, S. Merchant, A homolog of prokaryotic thiol disulfide transporter CcdA is required for the assembly of the cytochrome b6f complex in Arabidopsis chloroplasts, J. Biol. Chem. 279 (2004) 32474–32482.
[157] J.P. Jacquot, J. Vidal, P. Gadal, P. Schürmann, Evidence for the existence of several enzyme-specific thioredoxins in plants, FEBS Lett. 96 (1978) 243–246.
[158] V. Collin, E. Issakidis-Bourguet, C. Marchand, M. Hirasawa, J.M. Lancelin, D.B. Knaff, M. Miginiac-Maslow, The Arabidopsis plastidial thioredoxins: new functions and new insights into specificity, J. Biol. Chem. 278 (2003) 23747–23752.
[159] Y.H. Chi, J.C. Moon, J.H. Park, H.S. Kim, I.S. Zulfugarov, W.I. Fanata, H.H. Jang, J.R. Lee, Y.M. Lee, S.T. Kim, Y.Y. Chung, C.O. Lim, J.Y. Kim, D.J. Yun, C.H. Lee, K.O. Lee, S.Y. Lee, Abnormal chloroplast development and growth inhibition in rice thioredoxin m knock-down plants, Plant Physiol. 148 (2008) 808–817.
[160] T. Luo, T. Fan, Y. Liu, M. Rothbart, J. Yu, S. Zhou, B. Grimm, M. Luo, Thioredoxin redox regulates ATPase activity of magnesium chelatase CHLI subunit and modulates redox-mediated signaling in tetrapyrrole biosynthesis and homeostasis of reactive oxygen species in pea plants, Plant Physiol. 159 (2012) 118–130.
[161] P. Rey, R. Sanz-Barrio, G. Innocenti, B. Ksas, A. Courteille, D. Rumeau, E. Issakidis-Bourguet, I. Farran, Overexpression of plastidial thioredoxins f and m differentially alters photosynthetic activity and response to oxidative stress in tobacco plants, Front. Plant Sci. 4 (2013) 390.
[162] B.B. Buchanan, Y. Balmer, Redox regulation: a broadening horizon, Annu. Rev. Plant Biol. 56 (2005) 187–220.
[163] C. Job, L. Rajjou, Y. Lovigny, M. Belghazi, D. Job, Patterns of protein oxidation in Arabidopsis seeds and during germination, Plant Physiol. 138 (2005) 790–802.
[164] K. Kobrehel, B.C. Yee, B.B. Buchanan, Role of the NADP/thioredoxin system in the reduction of alpha-amylase and trypsin inhibitor proteins, J. Biol. Chem. 266 (1991) 16135–16140.
[165] M.J. Cho, J.H. Wong, C. Marx, W. Jiang, P.G. Lemaux, B.B. Buchanan, Overexpression of thioredoxin h leads to enhanced activity of starch debranching enzyme (pullulanase) in barley grain, Proc. Natl. Acad. Sci. U. S. A. 96 (1999) (14641-14546).
[166] J.H. Wong, Y.B. Kim, P.H. Ren, N. Cai, M.J. Cho, P. Hedden, P.G. Lemaux, B.B. Buchanan, Transgenic barley grain overexpressing thioredoxin shows evidence that the starchy endosperm communicates with the embryo and the aleurone, Proc. Natl. Acad. Sci. U. S. A. 99 (2002) 16325–16330.
[167] A.J. Serrato, J.M. Pérez-Ruiz, M.C. Spinola, F.J. Cejudo, A novel NADPH thioredoxin reductase, localized in the chloroplast, which deficiency causes hypersensitivity to abiotic stress in *Arabidopsis thaliana*, J. Biol. Chem. 279 (2004) 43821–43827.

[168] J.M. Pérez-Ruiz, M.C. Spínola, K. Kirchsteiger, J. Moreno, M. Sahrawy, F.J. Cejudo, Rice NTRC is a high-efficiency redox system for chloroplast protection against oxidative damage, Plant Cell 18 (2006) 2356–2368.
[169] A. Lepisto, S. Kangasjarvi, E.M. Luomala, G. Brader, N. Sipari, M. Keränen, M. Keinänen, E. Rintamäki, Chloroplast NADPH-thioredoxin reductase interacts with photoperiodic development in Arabidopsis, Plant Physiol. 149 (2009) 1261–2176.
[170] K. Kirchsteiger, J. Ferrandez, M.B. Pascual, M. Gonzalez, F.J. Cejudo, NADPH thioredoxin reductase C is localized in plastids of photosynthetic and nonphotosynthetic tissues and is involved in lateral root formation in Arabidopsis, Plant Cell 24 (2012) 1534–1548.
[171] J. Toivola, L. Nikkanen, K.M. Dahlstrôm, T.A. Salminen, A. Lepistö, F. Vignols, E. Rintamäki, Overexpression of chloroplast NADPH-dependent thioredoxin reductase in Arabidopsis enhances leaf growth and elucidates in vivo function of reductase and thioredoxin domains, Front. Plant Sci. 4 (2013) 389.
[172] A.S. Richter, E. Peter, M. Rothbart, M. Schlicke, H. Schlicke, J. Toivola, E. Rintamäki, B. Grimm, Posttranslational influence of NADPH-dependent thioredoxin reductase C on enzymes in tetrapyrrole synthesis, Plant Physiol. 162 (2013) 63–73.
[173] J. Michalska, H. Zauber, B.B. Buchanan, F.J. Cejudo, P. Geigenberger, NTRC links built-in thioredoxin to light and sucrose in regulating starch synthesis in chloroplasts and amyloplasts, Proc. Natl. Acad. Sci. U. S. A. 106 (2009) 9908–9913.
[174] A. Lepisto, E. Paluka, J. Toivola, A. Krieger-Liszkay, F. Vignols, E. Rintamäki, Deletion of chloroplast NADPH-dependent thioredoxin reductase results in inability to regulate starch synthesis and causes stunted growth under short-day photoperiods, J. Exp. Bot. 64 (2013) 3848–3854.
[175] H.B. Chae, J.C. Moon, M.R. Shin, Y.H. Chi, Y.J. Jung, S.Y. Lee, G.M. Nawkar, H.S. Jung, J.K. Hyun, W.Y. Kim, C.H. Kang, D.J. Yun, K.O. Lee, S.Y. Lee, Thioredoxin reductase type C (NTRC) orchestrates enhanced thermotolerance to Arabidopsis by its redox-dependent holdase chaperone function, Mol. Plant 6 (2013) 323–336.
[176] I. Thormählen, J. Ruber, E. von Roepenack-Lahaye, S.M. Ehrlich, V. Massot, C. Hümmer, J. Tezycka, E. Issakidis-Bourguet, P. Geigenberger, Inactivation of thioredoxin f1 leads to decreased light activation of ADP-glucose pyrophosphorylase and altered diurnal starch turnover in leaves of Arabidopsis plants, Plant Cell Environ. 36 (2013) 16–29.
[177] R. Sanz-Barrio, P. Corral-Martinez, M. Ancin, J.M. Segui-Simarro, I. Farran, Overexpression of plastidial thioredoxin f leads to enhanced starch accumulation in tobacco leaves, Plant Biotechnol. J. 11 (2013) 618–627.
[178] S.D. Clouse, J.M. Sasse, Brassinosteroids: essential regulators of plant growth and development, Annu. Rev. Plant Physiol. Plant Mol. Biol. 49 (1998) 427–451.
[179] Y.P. Jiang, F. Cheng, Y.H. Zhou, X.J. Xia, W.H. Mao, K. Shi, Z.X. Chen, J.Q. Yu, Cellular glutathione redox homeostasis plays an important role in the brassinosteroid-induced increase in $CO_2$ assimilation in *Cucumis sativus*, New Phytol. 194 (2012) 932–943.
[180] F. Cheng, Y.H. Zhou, X.J. Xia, K. Shi, J. Zhou, J.Q. Yu, Chloroplastic thioredoxin-f and thioredoxin-m1/4 play important roles in brassinosteroids-induced changes in $CO_2$ assimilation and cellular redox homeostasis in tomato, J. Exp. Bot. 65 (2014) 4335–4347.
[181] K. Apel, H. Hirt, Reactive oxygen species: metabolism, oxidative stress, and signal transduction, Annu. Rev. Plant Biol. 55 (2004) 373–399.
[182] K.J. Dietz, Peroxiredoxins in plants and cyanobacteria, Antioxid. Redox Signal. 15 (2011) 1129–1159.
[183] L. Tarrago, E. Laugier, P. Rey, Protein-repairing methionine sulfoxide reductases in photosynthetic organisms: gene organization, reduction mechanisms, and physiological roles, Mol. Plant 2 (2009) 202–217.
[184] M. Broin, S. Cuiné, F. Eymery, P. Rey, The plastidic 2-Cys peroxiredoxin is a target for a thioredoxin involved in the protection of the photosynthetic apparatus against oxidative damage, Plant Cell 14 (2002) 1417–1432.
[185] P. Rey, S. Cuiné, F. Eymery, J. Garin, M. Court, J.P. Jacquot, N. Rouhier, M. Broin, Analysis of the proteins targeted by CDSP32, a plastidic thioredoxin participating in oxidative stress responses, Plant J. 41 (2005) 31–42.
[186] L. Tarrago, E. Laugier, M. Zaffagnini, C. Marchand, P. Le Maréchal, S.D. Lemaire, P. Rey, Plant thioredoxin CDSP32 regenerates 1-Cys methionine sulfoxide reductase B activity through the direct reduction of sulfenic acid, J. Biol. Chem. 285 (2010) 14964–14972.
[187] P. Rey, G. Pruvot, N. Becuwe, F. Eymery, D. Rumeau, G. Peltier, A novel thioredoxin-like protein located in the chloroplast is induced by water deficit in *Solanum tuberosum* L. plants, Plant J. 13 (1998) 97–107.
[188] M. Broin, I. Besse, P. Rey, Evidence for post-translational control in the expression of a gene encoding a plastidic thioredoxin during leaf development in *Solanum tuberosum* plants, Plant Physiol. Biochem. 41 (2003) 303–308.
[189] C. Bowler, M. van Montagu, D. Inzé, Superoxide dismutase and stress tolerance, Annu. Rev. Plant Physiol. Plant Mol. Biol. 43 (1992) 83–116.
[190] M. Havaux, J.P. Bonfils, C. Lütz, K.K. Niyogi, Photodamage of the photosynthetic apparatus and its dependence on the leaf developmental stage in the *npq1* Arabidopsis mutant deficient in the xanthophyll cycle enzyme violaxanthin de-epoxidase, Plant Physiol. 124 (2000) 273–284.
[191] V. Collin, P. Lamkemeyer, M. Miginiac-Maslow, M. Hirasawa, D.B. Knaff, K.J. Dietz, E. Issakidis-Bourguet, Characterization of plastidial thioredoxins from Arabidopsis belonging to the new y-type, Plant Physiol. 136 (2004) 4088–4095.
[192] N. Navrot, V. Collin, J. Gualberto, E. Gelhaye, M. Hirasawa, P. Rey, D.B. Knaff, E. Issakidis, J.P. Jacquot, N. Rouhier, Plant glutathione peroxidases are functional peroxiredoxins distributed in several subcellular compartments and regulated during biotic and abiotic stresses, Plant Physiol. 142 (2006) 1364–1379.
[193] C. Vieira Dos Santos, E. Laugier, L. Tarrago, V. Massot, E. Issakidis-Bourguet, N. Rouhier, P. Rey, Specificity of thioredoxins and glutaredoxins as electron donors to two distinct classes of Arabidopsis plastidial methionine sulfoxide reductases B, FEBS Lett. 581 (2007) 4371–4376.
[194] E. Laugier, L. Tarrago, A. Courteille, G. Innocenti, F. Eymery, D. Rumeau, E. Issakidis-Bourguet, P. Rey, Involvement of thioredoxin y2 in the preservation of leaf methionine sulfoxide reductase capacity and growth under high light, Plant Cell Environ. 36 (2013) 670–682.
[195] E. Châtelain, P. Satour, E. Laugier, B. Ly Vu, N. Payet, P. Rey, F. Montrichard, Evidence for the participation of the methionine sulfoxide reductase repair system in plant seed longevity, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 3633–3638.
[196] J.H. Schippers, H.M. Nguyen, D. Lu, R. Schmidt, B. Mueller-Roeber, ROS homeostasis during development: an evolutionary conserved strategy, Cell. Mol. Life Sci. 69 (2012) 3245–3257.
[197] H. Tsukagoshi, W. Busch, P.N. Benfey, Transcriptional regulation of ROS controls transition from proliferation to differentiation in the root, Cell 143 (2010) 606–616.
[198] J. Foreman, V. Demidchik, J.H. Bothwell, P. Mylona, H. Miedema, M.A. Torres, P. Linstead, S. Costa, C. Brownlee, J.D. Jones, J.M. Davies, L. Dolan, Reactive oxygen species produced by NADPH oxidase regulate plant cell growth, Nature 422 (2003) 442–446.
[199] K. Sundaravelpandian, N.N. Chandrika, W. Schmidt, PFT1, a transcriptional Mediator complex subunit, controls root hair differentiation through reactive oxygen species (ROS) distribution in Arabidopsis, New Phytol. 197 (2013) 151–161.
[200] N. Rouhier, J.P. Jacquot, The plant multigenic family of thiol peroxidases, Free Radic. Biol. Med. 38 (2005) 1413–1421.
[201] R.A.P. Stacy, E. Munthe, T. Steinum, B. Sharma, R.B. Aalen, A peroxiredoxin antioxidant is encoded by a dormancy-related gene, Per1, expressed during late development in aleurone and embryo of barley grains, Plant Mol. Biol. 31 (1996) 1205–1216.
[202] K.O. Lee, H.H. Jang, B.G. Jung, Y.H. Chi, J.Y. Lee, Y.O. Choi, J.R. Lee, C.O. Lim, M.J. Cho, S.Y. Lee, Rice 1Cys-peroxiredoxin over-expressed in transgenic tobacco does not maintain dormancy but enhances antioxidant activity, FEBS Lett. 486 (2000) 103–106.
[203] C. Haslekas, M.K. Viken, P.E. Grini, V. Nygaard, S.H. Nordgard, T.J. Meza, R.B. Aalen, Seed 1-Cystein peroxiredoxin antioxidants are not involved in dormancy, but contribute to inhibition of germination during stress, Plant Physiol. 133 (2003) 1148–1157.
[204] P. Pulido, R. Cazalis, F.J. Cejudo, An antioxidant redox system in the nucleus of wheat seed cells suffering oxidative stress, Plant J. 57 (2009) 132–145.
[205] C. Bréhelin, E.H. Meyer, J.P. de Souris, G. Bonnard, Y. Meyer, Resemblance and dissemblance of Arabidopsis type II peroxiredoxins: similar sequences for divergent gene expression, protein localization and activity, Plant Physiol. 132 (2003) 2045–2057.
[206] I. Finkemeier, M. Goodman, P. Lankemeyer, A. Kandlbinder, L.J. Sweetlove, K.J. Dietz, The mitochondrial type II peroxiredoxin F is essential for redox homeostasis and root growth of *Arabidopsis thaliana* under stress, J. Biol. Chem. 280 (2005) 12168–12180.
[207] L. Haluskova, K. Valentovicova, J. Huttova, I. Mistrik, L. Tamas, Effect of abiotic stresses on glutathione peroxidase and glutathione S-transferase activity in barley root tips, Plant Physiol. Biochem. 47 (2009) 1069–1074.
[208] G. Passaia, L.S. Fonini, A. Caverzan, D. Jardim-Messeder, A.P. Christoff, M.L. Gaeta, J.E. de Araujo Mariath, R. Margis, M. Margis-Pinheiro, The mitochondrial glutathione peroxidase GPX3 is essential for $H_2O_2$ homeostasis and root and shoot development in rice, Plant Sci. 208 (2013) 93–101.
[209] G. Passaia, G. Queval, J. Bai, M. Margis-Pinheiro, C.H. Foyer, The effects of redox controls mediated by glutathione peroxidases on root architecture in *Arabidopsis thaliana*, J. Exp. Bot. 65 (2014) 1403–1413.
[210] C.C. Chang, I. Slesak, L. Jordá, A. Sotnikov, M. Melzer, Z. Miszalski, P.M. Mullineaux, J.E. Parker, B. Karpinska, S. Karpinski, Arabidopsis chloroplastic glutathione peroxidases play a role in cross talk between photooxidative stress and immune responses, Plant Physiol. 150 (2009) 670–683.
[211] G.C. Pagnussat, H.J. Yu, Q.A. Ngo, S. Rajani, S. Mayalagu, C.S. Johnson, A. Capron, L.F. Xie, D. Ye, V. Sundaresan, Genetic and molecular identification of genes required for female gametophyte development and function in Arabidopsis, Development 132 (2005) 603–614.
[212] Y. Miao, D. Lv, P. Wang, X.C. Wang, J. Chen, C. Miao, C.P. Song, An Arabidopsis glutathione peroxidase functions as both a redox transducer and a scavenger in abscisic acid and drought stress responses, Plant Cell 18 (2006) 2749–2766.
[213] U. Bechtold, D.J. Murphy, P.M. Mullineaux, Arabidopsis peptide methionine sulfoxide reductase2 prevents cellular oxidative damage in long nights, Plant Cell 16 (2004) 908–919.
[214] E. Laugier, L. Tarrago, C. Vieira Dos Santos, F. Eymery, M. Havaux, P. Rey, *Arabidopsis thaliana* plastidial methionine sulfoxide reductases B, MSRBs, account for most leaf peptide MSR activity and are essential for growth under environmental constraints through a role in the preservation of photosystem antennae, Plant J. 61 (2010) 271–282.
[215] C.W. Li, S.H. Lee, P.S. Chieh, C.S. Lin, Y.C. Wang, M.T. Chan, Arabidopsis root-abundant cytosolic methionine sulfoxide reductase B genes MsrB7 and MsrB8 are involved in tolerance to oxidative stress, Plant Cell Physiol. 53 (2012) 1707–1719.
[216] S.K. Oh, K.H. Baek, E.S. Seong, Y.H. Joung, G.J. Choi, J.M. Park, H.S. Cho, E.A. Kim, S. Lee, D. Choi, CaMsrB2, pepper methionine sulfoxide reductase B2, is a novel defense regulator against oxidative stress and pathogen attack, Plant Physiol. 154 (2010) 245–261.
[217] N.P. Hüner, R. Bode, K. Dahal, L. Hollis, D. Rosso, M. Krol, A.G. Ivanov, Chloroplast redox imbalance governs phenotypic plasticity: the "grand design of photosynthesis" revisited, Front. Plant Sci. 3 (2012) 255.

[218] I.M. Møller, P.E. Jensen, A. Hansson, Oxidative modifications to cellular components in plants, Annu. Rev. Plant Biol. 58 (2007) 459–481.
[219] M.J. Davies, The oxidative environment and protein damage, Biochim. Biophys. Acta 1703 (2005) 93–109.
[220] Y.J. Liu, A. Nunes-Nesi, S.V. Wallström, I. Lager, A.M. Michalecka, F.E.B. Norberg, S. Widell, K.M. Fredlund, A.R. Fernie, A.G. Rasmusson, A redox-mediated modulation of stem bolting in transgenic *Nicotiana sylvestris* differentially expressing the external mitochondrial NADPH dehydrogenase, Plant Physiol. 150 (2009) 1248–1259.
[221] G. Kocsy, I. Tari, R. Vanková, B. Zechmann, Z. Gulyás, P. Poór, G. Galiba, Redox control of plant growth and development, Plant Sci. 211 (2013) 77–91.
[222] M.J. Considine, C.H. Foyer, Redox regulation of plant development, Antioxid. Redox Signal. 21 (2014) 1305–1326.
[223] C.H. Foyer, G. Noctor, Redox signaling in plants, Antioxid. Redox Signal. 18 (2013) 2087–2090.
[224] B. Morgan, M.C. Sobotta, T.P. Dick, Measuring E(GSH) and $H_2O_2$ with roGFP2-based redox probes, Free Radic. Biol. Med. 51 (2011) 1943–1951.
[225] G. Brunoud, D.M. Wells, M. Oliva, A. Larrieu, V. Mirabet, A.H. Burrow, T. Beeckman, S. Kepinski, J. Traas, M.J. Bennett, T. Vernoux, A novel sensor to map auxin response and distribution at high spatio-temporal resolution, Nature 482 (2012) 103–106.
[226] N. Rouhier, A. Villarejo, M. Srivastava, E. Gelhaye, O. Keech, M. Droux, I. Finkemeier, G. Samuelsson, K.J. Dietz, J.P. Jacquot, G. Wingsle, Identification of plant glutaredoxin targets, Antioxid. Redox Signal. 7 (2005) 919–929.
[227] T.P. Howard, M.J. Fryer, P. Singh, M. Metodiev, A. Lytovchenko, T. Obata, A.R. Fernie, N.J. Kruger, W.P. Quick, J.C. Lloyd, C.A. Raines, Antisense suppression of the small chloroplast protein CP12 in tobacco alters carbon partitioning and severely restricts growth, Plant Physiol. 157 (2011) 620–631.
[228] M.C. Espunya, M. Diaz, J. Moreno-Romero, M.C. Martinez, Modification of intracellular levels of glutathione-dependent formaldehyde dehydrogenase alters glutathione homeostasis and root development, Plant Cell Environ. 29 (2006) 1002–1011.
[229] S. Xu, D. Guerra, U. Lee, E. Vierling, S-nitrosoglutathione reductases are low-copy number, cysteine-rich proteins in plants that control multiple developmental and defense responses in Arabidopsis, Front. Plant Sci. 4 (2013) 430.
[230] U. Lee, C. Wie, B.O. Fernandez, M. Feelisch, E. Vierling, Modulation of nitrosative stress by S-nitrosoglutathione reductase is critical for thermotolerance and plant growth in Arabidopsis, Plant Cell 20 (2008) 786–802.
[231] E. Kwon, A. Feechan, B.W. Yun, B.H. Hwang, J.A. Pallas, J.G. Kang, G.J. Loake, AtGSNOR1 function is required for multiple developmental programs in Arabidopsis, Planta 236 (2012) 887–900.