University of South Bohemia in České Budějovice
Faculty of Science

The role of chromatin modifiers in plant response to light acclimation

Bachelor's thesis

Abdoulie Jallow

Supervisor: Mingxi Zhou Ph.D.
Co-supervisor: Mgr. Iva Mozgová Ph.D. (Principal Investigator)

České Budějovice 2023

Jallow, A., 2023: The role of chromatin modifiers in plant response to light acclimation. Bc. Thesis, in English. – 67 p., Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic.

Annotation

The primary objective of this thesis is to enhance our understanding of chromatin contribution to light signalling by screening for the *gun* (*genomes-uncoupled*) phenotype in transgenic plant lines carrying impaired chromatin modifiers. To achieve this, the study used RT-qPCR to analyse gene expression in mutant plants under chloroplast-damaging conditions. These expression profiles were then compared with those of wild-type plants under similar conditions.

DECLATATION

I declare that I am the author of this qualification thesis and that in writing it I have used the sources and literature displayed in the list of used sources only.

Linz, 04. 12. 2023

Acknowledgments

I extend my deepest gratitude to my supervisors, Dr. Mingxi Zhou and Dr. Iva Mozgová, for the opportunity to pursue this research and their invaluable mentorship. My thanks also to the members of Dr. Mozgová's lab for their supportive environment. Lastly, immense gratitude to my family and friends for their unwavering support.

Table of Contents

1. Introduction

Being sessile organisms, plants face various environmental stresses, including unpredictable fluctuations in sunlight intensity, which can exert both positive and negative effects on their growth and survival. To cope with light and other abiotic factors, plants have evolved a range of sophisticated sensing and response mechanisms at different levels, enabling them to fine-tune their photosynthetic processes and other physiological responses in an adaptive manner (Szymańska et al., 2017). One example of these adaptations is the regulation of stomatal movement. Stomata are microscopic pores on the leaf surface that control transpiration and gas exchange. By opening and closing their stomata, plants balance their water loss and carbon uptake.

Plants are among the life forms capable of synthesizing their own food utilizing energy from sunlight through a process called photosynthesis. Plants possess the green pigment chlorophyll, primarily located within the leaves, which facilitates energy harvesting to fuel the conversion of water and carbon dioxide into organic molecules. Plants also store excess carbon and energy in the form of starch, proteins or lipids in various parts of their body, including leaves, stems, roots, fruits, or seeds, which serve as a primary source of energy and carbon for humans and other animals. Moreover, plants play a critical role in maintaining the delicate balance of an ecosystem by releasing oxygen, recycling carbon dioxide, contributing to the mitigation of the greenhouse effect and climate change, and purifying the air, water, and soil in their immediate environment. Therefore, plants are indispensable for sustaining most of the biological processes that underpin life on Earth.

*Arabidopsis thaliana (*further Arabidopsis*)* emerged as a suitable model organism in plant research in the 1980s, supplanting previously used species such as maize, soybean, petunia, tomato, pea, and snapdragon (Koornneef & Meinke, 2010). The main rationale for choosing Arabidopsis as a model organism was its suitability for genetic experiments, due to its short life cycle, small size that conserves space, and abundant seed production through self-pollination (Koornneef & Meinke, 2010).

Plants exemplify primary endosymbiosis, which is characterised by the incorporation of an endosymbiotic prokaryote into a host cells, establishing a symbiotic relationship (Stadnichuk & Kusnetsov, 2021). During this evolutionary process, the internalised prokaryote undergoes

extensive evolutionary modifications and by losing a subset of genes that are transferred to the nucleus partially loses its autonomy, eventually transforming into an organelle that confers novel functional capabilities and advantages on the host eukaryotic cell (Archibald, 2015). This phenomenon explains the origin of two critical organelles in eukaryotic organisms: mitochondria and chloroplasts (Gray et al., 1999; Margulis, 1975). Mitochondria, commonly recognised as the 'powerhouse' of the cell, are responsible for the catabolism of fuel molecules and the generation of energy through a process called cellular respiration (Alberts et al., 2002). Chloroplasts serve as photosynthetic machinery in plants and algae, capturing light energy to synthesise sugars (Van Dingenen et al., 2016). Beyond their primary functions, both organelles are intricately involved in various cellular processes, including signaling pathways, stress response mechanisms, and the biosynthesis of a wide range of compounds (Jarvis & López-Juez, 2013). A key organelle in the plant cell is the nucleus, responsible for storing the DNA, representing majority of genetic material of the cell , and for regulating gene expression or coordinating a multitude of cellular activities (Zidovska, 2020). Nuclear DNA is assembled in a highly organised but dynamic structure known as chromatin, which controls DNA accessibility and expression of genes (Zidovska, 2020). Within chromatin, DNA is wrapped around octamers of basic proteins called histones, forming the nucleosome, or basic unit of chromatin. The openness or compaction of chromatin structure dictates the accessibility of DNA to the transcriptional apparatus, thereby regulating the expression of genes involved in a wide range of cellular processes, including genes that contribute to chloroplast functionality (Gibney & Nolan, 2010). Changes to chromatin structure rely on chemical modifications to DNA and histones that are read by chromatin remodelling proteins. These then alter DNA accessibility and thereby change, gene expression, without changing the underlying DNA sequence. This mode of gene transcription regulation is called “epigenetic” as it does not involve a change to genetic information.

Endosymbiosis has given rise to chloroplasts and mitochondria with semi-autonomous genomes that are remnants of the original genomes of the engulfed prokaryotic cell (Archibald, 2015). The chloroplast genome encodes for many proteins that are involved in photosynthesis as well as proteins that participate in the biosynthesis of many compounds (Song et al., 2021). However, the majority of proteins involved in photosynthesis and those that regulate plastid genome transcription are encoded by the nuclear genome and subsequently transported as proteins into the chloroplast (Sun & Zerges, 2015; Dobrogojski et al., 2020). This dual encoding of proteins necessitates a high level of coordination in gene expression between the

nuclear and plastid genomes (Wu & Bock, 2021). For instance, damage to chloroplasts often leads to a reduction in the transcription of photosynthesis-associated nuclear genes (PhANGs), demonstrating the intricate communication between these organelles (Inaba et al., 2011; Chi et al., 2013; Hills et al., 2015). Signals transmitted from the chloroplast to regulate the expression of nuclear genes within this communication loop are called retrograde signals (Richter et al., 2023). Conversely, signals emanating from the nucleus to regulate plastid gene expression (PGE) are referred to as anterograde signals (Woodson & Chory, 2008; Wu & Bock, 2021; Jan et al., 2022).

Despite advancements, the precise mechanisms underpinning this coordination remain enigmatic. Major progress in understanding the coordination of gene expression between nuclear and plastid genomes have been achieved through the identification of genome-uncoupled (*gun*) mutants. The seminal study by Susek et al. (1993) in *Arabidopsis thaliana* identified six *gun* mutants that disrupt the retrograde signaling pathway. Unlike wild-type plants, these mutants did not suppress the transcription of the nuclear-encoded gene *LIGHT-HARVESTING CHLOROPHYLL B-BINDING 1.2 (LHCB1.2)* following chloroplast damage, typifying the *gun* phenotype (Mochizuki et al., 1996; Susek et al., 1993; Wu & Bock, 2021; Richter et al., 2023). Five of the six identified mutations affected genes involved in tetrapyrrole metabolism, leading to a proposed role of tetrapyrroles in mediating the retrograde signal (Richter et al., 2023; Wu & Bock, 2021). How gene response is regulated in the nucleus, at the level of chromatin structure and transcriptional regulation, is even less understood.

Therefore, this thesis aims to extend our understanding of retrograde signaling by screening for the *gun* phenotype in plant lines carrying defective chromatin modifiers. This approach may uncover new mechanisms by which plants adjust to varying light conditions, further elucidating the complex interplay between nuclear and plastid genomes.

2. Literature review

2.1. Chloroplast development

Chloroplasts probably evolved from cyanobacterial endosymbionts that were engulfed by phagocytosis by a eukaryotic cell 1.5 billion years ago (Sabater, 2018; Yoon et al., 2004). Over time, most of its DNA was lost and only a small fraction of the original cyanobacterial genes remained in modern chloroplasts (Archibald, 2015). Chloroplasts enable plants to perform photosynthesis, the process of converting light energy into chemical energy. In addition to this primary role, chloroplasts are also involved in various metabolic pathways that produce or modify amino acids, pigments, fatty acids, plant hormones and other molecules (Cackett et al., 2022). Moreover, chloroplasts play a key role in plant adaptation to environmental stress by regulating the expression of stress-responsive genes and modulating the redox state of the cell (Littlejohn et al., 2021; Mamaeva et al., 2020; Spetea et al., 2014; Witte & Herde, 2020)

2.2. Chloroplast biogenesis

Plastids are not formed from scratch, but are inherited from parental progenitor cells (Pyke, 2007). Chloroplasts develop from small undifferentiated proplastids that subsequently undergo multiple rounds of division (chloroplast biogenesis). Other types of plastids are chromoplasts, amyloplasts, and etioplasts, which develop from the same proplastid that is inherited between parental and progeny cells and through generations. Based on condition and cell type, plastids can convert into other types. For instance. amyloplasts store starch and are especially found in starch accumulating tissues/organs, such as storage roots or tubers.

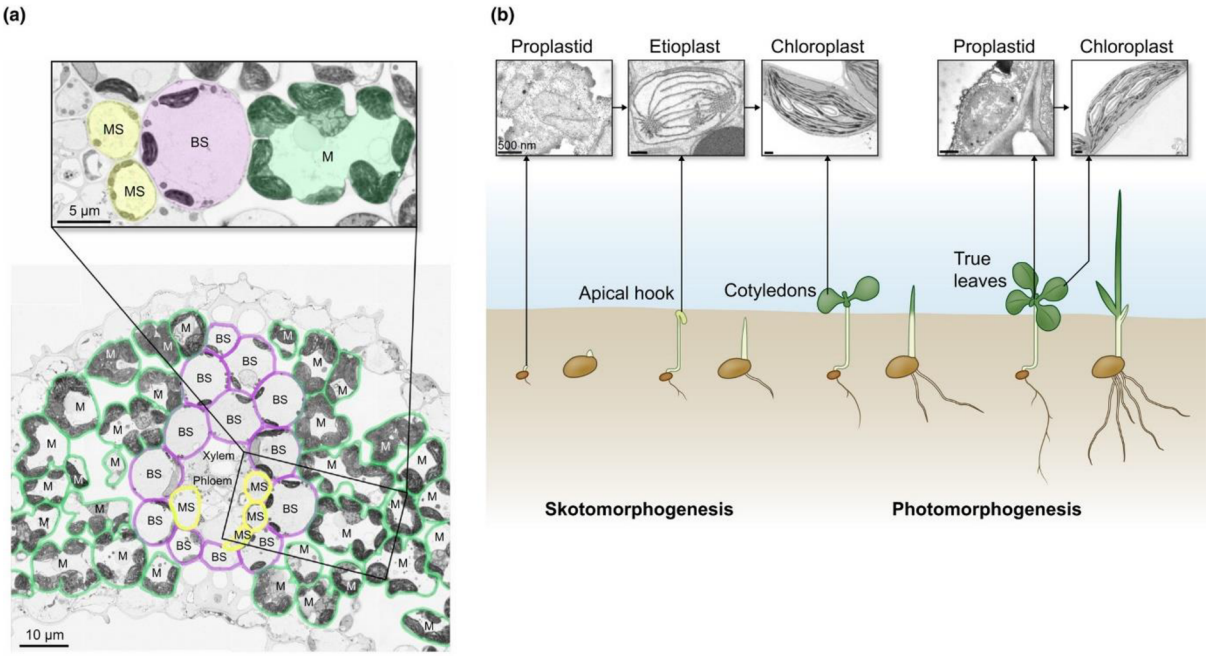

Figure 1: Chloroplast development in cotyledons and true leaves. (a) Despite all cells of the leaf receiving light, the chloroplast compartment varies between cell types. Mesophyll cells (M) contain many large chloroplasts whilst chloroplast occupancy of the bundle sheath (BS) and mestome sheath (MS) is lower. Image of transverse section of a rice leaf taken with transmission electron microscopy. Green, purple, and yellow colours indicate the M, BS and MS, respectively. (b) Schematic of skotomorphogenesis and photomorphogenesis in dicotyledons (with epigeal germination) and monocotyledons with representative images illustrating differentiation of plastids to chloroplasts during these processes in Arabidopsis. Adapted from "Chloroplast development in green plant tissues: the interplay between light, hormone, and transcriptional regulation" by Lee Cackett, 2021, New Phytologist, Volume: 233, Issue: 5, Pages: 2000-2016.

Skotomorphogenesis or etiolation is a developmental program that is activated when seeds begin to germinate in the dark and is characterized by rapid elongation of the hypocotyl (Cackett et al., 2022; Pipitone et al., n.d.). During skotomorphogenesis, proplastids undergo proliferation and differentiation to etioplasts within cotyledon cells. The inner membrane of the etioplast has a paracrystalline lattice (prolamellar body) and disk-shaped membranes, called prothylakoids (Floris & Kühlbrandt, 2021). The prolamellar body is the precursor structure of the photosynthetic machinery and consists of regular arrangements of NADPH, protochlorophyllide-oxido-reductase, chlorophyll precursor protochlorophyllide, thylakoid

membrane lipids digalactosyl-diacylglycerol (DGDG), and monogalactosyl-diacylglycerol (Bastien et al., 2016).

Photomorphogenesis begins when the seedling emerges from the soil and encounters light, causing rapid transformation of the etioplasts of the cotyledons into functional chloroplasts (Jarvis & López-Juez, 2013; Solymosi & Schoefs, 2010). The initiation of photomorphogenesis is regulated by a network of photoreceptors, mainly phytochromes, which activate the transcription of numerous nuclear genes encoding chloroplast proteins (Arsovski et al., 2012). Thylakoid biogenesis is a crucial step in chloroplast development since these internal membranes harbor the photosynthetic electron transport chain. The formation of thylakoids and the initiation of photosynthetic activity depend on the simultaneous production and orderly integration of chlorophylls, lipids, and proteins across different compartments (Jarvis & López-Juez, 2013). The import of nuclear-encoded preproteins is mediated by chaperones that recognize chloroplast signals and by multimeric complexes of translocons of the outer and inner chloroplast membranes (TOC and TIC, respectively), which constitute the general chloroplast protein import machinery, allowing selective import by specifically recognizing transit peptide sequences through TOC receptors (Day & Theg, 2018; Richardson & Schnell, 2020). Subsequently, imported chlorophyll-binding proteins need to be assembled with the light-absorbing cofactor chlorophyll. Final steps of chlorophyll biosynthesis and greening involve light-dependent activation of protochlorophyllide oxidoreductase, an enzyme that mediates the transformation of protochlorophyllide into chlorophyllides a and b, which is finally converted to chlorophyll a and b (Liebers et al., 2017). Therefore, finally, the fully assembled thylakoid membrane consists of Photosystem II, the cytochrome b_6f complex, Photosystem I and ATP synthase (Cackett et al., 2022).

2.3. Nuclear- and chloroplast-encoded proteins control chloroplast development

Modern plastids have a compact genome consisting of 50-200 protein-coding genes, although the exact number varies between species (Martin et al., 1998; Ponce-Toledo et al., 2019; Wu & Bock, 2021). However, the majority of plastid proteins, approximately 3000, are encoded by the nuclear genome (Christian et al., 2020; Richly et al., 2003). Therefore, proper development of chloroplasts and thus photosynthetic machinery depends on close coordination between the nuclear and chloroplast genomes, achieved by anterograde and retrograde signaling between the two organelles (Koussevitzky et al., 2007; Shimizu et al., 2019; Wu & Bock, 2021). PGE is regulated by anterograde signals originating from the nucleus (Jan et al., 2022; Jarvis &

López-Juez, 2013; Shimizu et al., 2019). In turn, chloroplast development can affect nuclear gene expression through retrograde signaling (Chan et al., 2016; de Souza et al., 2017; Gläßer et al., 2014; Woodson & Chory, 2008).

Nuclear-encoded proteins are involved in a wide range of functions, including import processes, thylakoid development, protein maturation and degradation, plastid gene expression, chlorophyll synthesis, RNA processing, metabolite transport, and photosystem assembly (Waters & Langdale, 2009). The bulk of nuclear-encoded chloroplast-targeted proteins are initially synthesized by ribosomes in the cytosol as preproteins and are subsequently imported into the chloroplast. In most cases, pre-proteins possess transit peptides at their N-terminus, which facilitate their translocation into the chloroplast via interactions with TOC and TIC (Sjuts et al., 2017). The import process is facilitated by chloroplast heat shock proteins, which provide the necessary energy, and the subsequent processing of pre-proteins occurs through the action of a stromal processing peptidase (Sjuts et al., 2017).

The small chloroplast genome encodes mostly proteins involve in photosynthesis, transcription and translation (Daniell et al., 2016). Plastid transcription involves two types of polymerases: nuclear-encoded polymerases (NEPs) and plastid-encoded polymerases (PEPs) (Börner et al., 2015; Hedtke et al., 1997). The initiation of PEP-mediated transcription of chloroplast genes is regulated by a group of plastidic SIGMA (SIGs) factors (Chi et al., 2015). The transcription and translation of these chloroplast genes are crucial for the development of chloroplasts. For instance, mutant alleles associated with the PEP complexes exhibit an albino phenotype (Yuan et al., 2019). Similarly, *sig6* mutants, due to their inability to produce adequate chloroplast-encoded proteins, show deficiencies in chloroplast biogenesis (Chi et al., 2010). Moreover, mutants that are completely defective in NEP result in embryo lethality (Cackett et al., 2022; Hricová et al., 2006).

2.4. Retrograde signalling controls chloroplast and PhANG transcription

To date, five distinct classes of retrograde signals have been identified:1) plastid pigments or their precursors (tetrapyrroles, apocarotenoids, and carotenoids); 2) signals originating from plastid gene expression (PGE); 3) reactive oxygen species (ROS); 4) redox signals related to photosynthesis; and 5) changes in metabolite pools or fluxes (Chan et al., 2016; de Souza et al., 2017; Hernández-Verdeja & Strand, 2018). These signals manifest at different times and in different developmental contexts, and certain signals can exhibit varied effects under different conditions (Liebers et al., 2022). Therefore, identified retrograde signals can be further grouped

into two classes: “biogenic control” signals that informs the nucleus of the developmental status of the plastid and protein demand during early chloroplast formation, germination, and seedling growth (Liebers et al., 2022; Richter et al., 2023), and “operational control” signals generated by mature chloroplasts in response to environmental stimuli that changes the demand for nuclear-encoded proteins in plastids (Barajas-López et al., 2013; Hernández-Verdeja & Strand, 2018).

2.4.1. Biogenic retrograding signaling

Chloroplast biogenesis requires the precise integration of photosynthetic complexes that mediate electron transfer reactions during photosynthesis (Jarvis & López-Juez, 2013). This process involves the coordinated expression and assembly of protein subunits encoded by both plastid and nuclear genomes as well as the simultaneous incorporation of photosynthetic pigments (chlorophylls and carotenoids) and redox-active cofactors (e.g. hemes and iron–sulfur clusters) (Wu & Bock, 2021).

by Susek The pioneering study et al. (1993), which identified the *gun* (*genome-uncoupled*) mutants, marked the beginning of efforts to unravel the mechanisms of retrograde signaling in plants. The screening conducted by Susek et al. (1993) was based on the observation that the application of specific chemicals, such as the herbicide norflurazon (NF), which impairs chloroplast carotenoid biosynthesis, or inhibitors of organellar protein synthesis, such as lincomycin (LIN), lead to reduced levels of transcripts derived from photosynthesis-associated nuclear genes (PhANGs), particularly the *LHCB* marker gene encoding light-harvesting chlorophyll a/b-binding proteins of photosystem II (Mulo et al., 2003a; Oelmüller et al., 1986). A *gun* phenotype arises when seedlings continue to express PhANGs despite blockage of plastid development by NF.

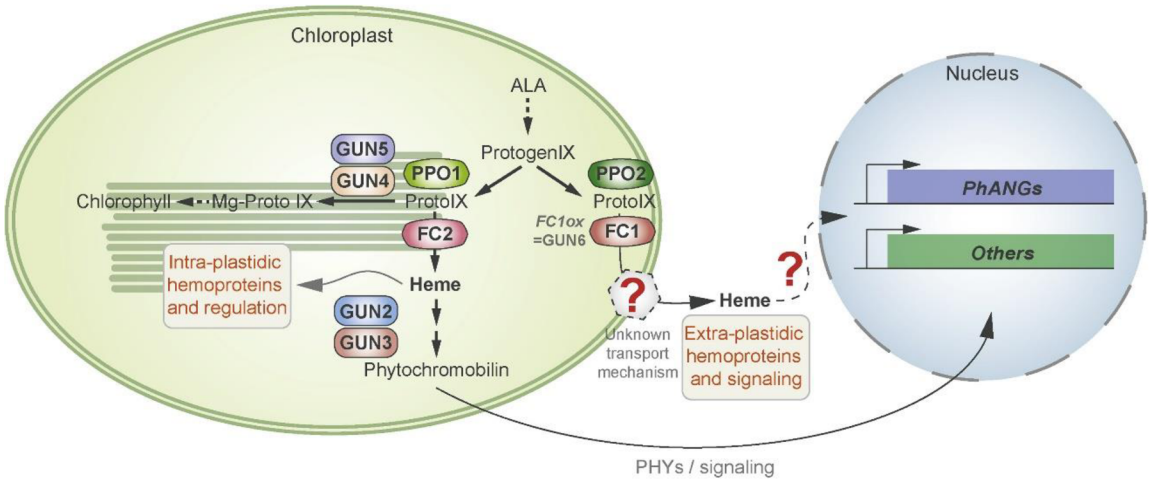

Figure 2: The tetrapyrrole pathway transforms glutamate into 5-aminolevulinic acid, a rate-limiting step that requires eight 5-aminolevulinic acid molecules to generate porphyrins. Protoporphyrin IX serves as a substrate for the two metal-chelating chelatases, Mg chelatase (MgCh) and ferrochelatase (FC), which branch at the point of chlorophyll and heme synthesis. The MgCh complex is composed of the subunits CHLH/GUN5, CHLI1, CHLI2, and CHLD, with GUN4 acting as a regulator. The isoforms PPO1 and FC2 are presumed to supply the heme used in plastids, which is further converted into phytochromobilin by the heme oxygenases HO1/GUN2, HO3, and HO4 and phytochromobilin synthase (HY2/GUN3). PPO2 and FC1/GUN6 are believed to produce the heme necessary for heme-dependent proteins outside of the plastids, with the heme pool speculated to function as a retrograde signal. A heme transporter is hypothesized to export heme into the cytoplasm. Notably, in A. thaliana, mitochondrial localization of PPO and FC can be ruled out, indicating that heme is exclusively produced in chloroplasts. Adapted from "Retrograde signaling in plants: A critical review focusing on the GUN pathway and beyond" by S. Richter, 2023, Plant communications, Volume 4, Issue: 1.

In the initial screen, five mutants (*gun 1–gun 5*) were isolated (Susek et al., 1993), and subsequent studies revealed that overexpression of plastid *FERROCHELATASE 1* (*FC1*) in *gun 6-1D* mutants also leads to a *gun* phenotype (Woodson et al., 2011). It should be noted that, except for GUN1, all the identified GUN proteins (GUN2 - GUN6) are directly involved in the biosynthesis of tetrapyrroles (TPB), either as enzymes or regulatory proteins within the pathway (Figure 2). GUN2, which is identical to HY1, functions as a heme oxygenase that converts heme to biliverdin iXα (Mochizuki et al., 2001). In contrast, GUN3 (or HY2), acts as

a phytochromobilin synthase, reducing biliverdin iXα to phytochromobilin (Kohchi et al., 2001; Mochizuki et al., 2001). GUN4 binds porphyrins and the ChlH subunit of Mg chelatase, thereby stimulating chlorophyll biosynthesis by activating Mg chelatase (Adhikari et al., 2011). GUN5, the ChlH subunit of Mg chelatase, is a key enzyme in the tetrapyrrole biosynthesis pathway involved in chlorophyll production (Mochizuki et al., 2001). Finally, the *gun6-1D* mutant, also known as *OeFC*, overexpresses plastid *FC1*, although not all *FC1* overexpression lines are considered *gun* mutants (Page et al., 2020; Woodson et al., 2011). These findings strongly indicate the crucial involvement of TPB-derived metabolites, and in particular heme, as retrograde signals in chloroplast biogenesis, (Z.-W. Zhang et al., 2015; Wu & Bock, 2021; A. S. Richter et al., 2023).

2.4.1.1. GUN1: A Central Regulator of Plastid Retrograde Signalling

GUN1, the pentatricopeptide repeat (PPR) protein with a small MutS-related (SMR) domain has emerged as a central node in multiple plastid retrograde signaling pathways, coordinating the expression of plastid genome-encoded genes and integrating signals from various retrograde signaling pathways (Koussevitzky et al., 2007; Woodson et al., 2013; Wu, Meyer, Richter, et al., 2019).

The initiation of chloroplast gene transcription mediated by PEPs is controlled by SIGMA (SIGs) factors (Fu et al., 2021). Angiosperms have six groups of sigma factor genes (SIG1-SIG6) that are involved in chloroplast development and chlorophyll biosynthesis by regulating the transcription of photosynthetic genes and tRNAs (Chi et al., 2015; Zhang et al., 2015). SIG2 and SIG6 regulate *PhANGs* via a partially redundant but distinct pathway and are complemented by *gun1* (Woodson et al., 2013).

The molecular function of GUN1 has remained elusive because of its low protein levels (Y. Jia et al., 2019; Wu & Bock, 2021). Recent studies have revealed that GUN1 accumulates during active chloroplast biogenesis and is highly expressed in tissues undergoing active chloroplast development (Wu et al., 2018). Post-transcriptional mechanisms regulate GUN1 protein accumulation, with rapid degradation mediated by stromal Clp protease and an involvement of chloroplast chaperone ClpC1. GUN1 contains PPR tracts and an SMR domain, suggesting that it is involved in organellar gene expression and nucleic acid binding (Koussevitzky et al., 2007; Pesaresi & Kim, 2019; Wu et al., 2018). However, subsequent studies have shown that GUN1 interacts with proteins rather than with nucleic acids (Tadini et al., 2016; Wu et al., 2019).

GUN1 interacts with various proteins involved in plastid retrograde signalling, including enzymes of the tetrapyrrole biosynthesis (TPB) pathway and chaperones (L. Liu et al., 2014).

It has also been found recently, that GUN1 facilitates protein import under stress conditions, thereby maintaining plastid protein homeostasis (Wu, et al., 2019). Loss of GUN1 function leads to accumulation of unimported precursor proteins in the cytosol. This perturbation triggers the accumulation of cytosolic chaperones, particularly HSP90, which participate in both the TPB and PGE pathways of retrograde signalling (Wu et al., 2019; Wu & Bock, 2021). The binding of Mg-ProtoIX to HSP90 inhibits its ATPase activity, suggesting a negative signal from the stressed plastids (Kindgren et al., 2011; Wu et al., 2019). Proteomic analyses have revealed significant differences in the abundance of proteins between wild-type and *gun1* mutant plants, particularly under stressful conditions (Wu & Bock, 2021). GUN1 has therefore been implicated in regulating plastid proteostasis, facilitating the import of chaperones, and ensuring balanced plastid protein homeostasis under stressful conditions (Wu et al., 2019).

Overall, GUN1 plays a pivotal role in plastid retrograde signaling, protein import, and plastid proteostasis. Its function is conditional, as it acts as a modulator of protein import and proteostasis only when plastid homeostasis is disturbed. Disruptions in PGE and protein import processes contribute to the *gun1* mutant phenotype and hypersensitivity to stress. Further investigation is needed to fully elucidate the molecular mechanisms underlying the function of GUN1 in biogenic retrograde signaling and plastid protein homeostasis.

2.4.2. Retrograde signaling in mature chloroplast

Retrograde signaling in mature chloroplasts differs from biogenic signaling in that it aims to maintain chloroplast and cellular homeostasis under various environmental and developmental conditions that affect plastid function.

2.4.2.1. Tetrapyrrole biosynthesis pathway

Plants synthesize four major tetrapyrroles—chlorophyll, heme, siroheme, and phytochromobilin—via a common branched pathway in the plastids. The pathway originates from glutamate and splits into two branches that use either Mg^{2+} (for chlorophyll) or Fe^{2+} (for heme and bilins) as cofactors. It is regulated at multiple levels to prevent the accumulation of excess tetrapyrroles, which can generate harmful radicals when exposed to light (Kobayashi & Masuda, 2016). Changes in the pathway reflect developmental and environmental changes and can affect the expression of nuclear genes related to photosynthesis in both green algae and

plants. Thus, the tetrapyrrole biosynthesis pathway is linked to both biogenic and operational control.

The TBP flux is influenced by environmental changes, leading to reactive oxygen species (ROS) and specific intermediate metabolites accumulation. *gun5* mutants are more vulnerable to abiotic stresses such as low temperature exposure (Kindgren et al., 2015). The aerobic cyclase reaction, which is very sensitive to oxidative stress and ROS, was found to result in the accumulation of upstream intermediates such as MgProtoIX/MgProtoIX-methylester in Arabidopsis (Stenbaek et al., 2008). These intermediates may exit the chloroplast through an unknown transporter, interact with and inhibit the ATPase activity of an HSP90 chaperone complex in the cytosol, and then regulate specific nuclear gene expression through transcription factors HY5 and PSEUDO RESPONSE REGULATOR 5 (PRR5) via ZEITLUPE (ZTL) (Crawford et al., 2018; Kindgren et al., 2011; Norén et al., 2016).

2.4.2.2. Methylerythritol cyclodiphosphate

Methylerythritol cyclodiphosphate (MEcPP) is a metabolite that originates from plastids and acts as an operational retrograde signal to modulate nuclear gene expression (Xiao et al., 2012). MEcPP plays a pivotal role in influencing hypocotyl length under red light conditions by regulating phyB protein levels through a calcium-mediated pathway involving the transcription factor CALMODULIN-BINDING TRANSCRIPTIONAL ACTIVATOR 3 (CAMTA3) (Jiang et al., 2019). Studies indicate that MEcPP triggers the expression of stress-responsive genes located in the nucleus, including chloroplast-localized and endoplasmic reticulum (ER) stress-related proteins (de Souza et al., 2017). Furthermore, MEcPP functions as a retrograde signal, activating nuclear genes associated with stress response and biotic defense, such as the JA-related enzyme-encoding *HYDROPEROXIDE LYASE* (*HPL*) (Xiao et al., 2012). Excessive MEcPP accumulation in stressed plants hampers growth and induces early flowering via the involvement of B-BOX DOMAIN PROTEIN 19 (BBX19) (de Souza et al., 2017).

Recent research highlights that MEcPP serves as a retrograde signal, inducing the unfolded protein response (UPR) upon ER stress caused by misfolded proteins (J.-X. Liu & Howell, 2016). This UPR involves pathways mediated by INISITOL REQUIRING 1 (IRE1), basic leucine zipper 60 (bZIP60), binding immunoglobulin proteins (BiPs), and the TFs bZIP28, and bZIP17 (Bao & Howell, 2017). MEcPP activates UPR by directly prompting the expression of bZIP60, IRE1a, bZIP28, and BiP3 through the nuclear transcription factor CAMTA3 in a calcium-dependent manner (Crawford et al., 2018; Walley et al., 2015).

2.5. Role of transcription factors in chloroplast development

The differentiation and function of plastids during plant development are controlled by transcription factors from two families, GOLDEN2-LIKE (GLK1&2) and GATA (GNC and CGA1), which regulate the transcription of chloroplast-related genes (Bastakis et al., 2018).

GOLDEN2-LIKE (GLK1 and 2) was first discovered in maize (Langdale & Kidner, 1994; Hall et al., 1998). The GLK1 and GLK2 transcription factor genes are functionally redundant in Arabidopsis, and their mutants exhibit smaller chloroplasts with incorrectly formed thylakoids in bundle-sheath cells and reduced chlorophyll content. The expression of nuclear genes involved in light harvesting and chlorophyll biosynthesis was reduced in these mutants. These genes include the LIGHT HARVESTING COMPLEX (LHC) PROTEIN family and tetrapyrrole synthesis genes Glutamyl-tRNA reductase family protein (HEMA1), GUN4, GUN5/CHLH, and CAO (Fitter et al., 2002; Rossini et al., 2001; Waters et al., 2009). The molecular mechanisms underlying the pale green appearance (reduced chlorophyll content) of the *glk1 glk2* mutants are probably the impaired function of the light harvesting complex and reduced chlorophyll synthesis (Bastakis et al., 2018). Elevated expression of GLK transcription factors enhances chlorophyll synthesis and increases chloroplast production. This upregulation of GLKs has been associated with the promotion of chloroplast development, even in tissues that typically exhibit lower chloroplast content (Wang et al., 2017). Furthermore, overexpression of GLK1 or GLK2 has been observed to display *gun* phenotypes, indicating a disruption of the nuclear response to chloroplast-initiated retrograde signaling (Leister & Kleine, 2016).

The GATA transcription factor family comprises GNC and CGA1/GNL transcription factors, which were identified based on their robust induction in response to various environmental disturbances such as light and nitrate (Hudson et al., 2013). CGA1/GNL also appeared to have a broad influence on several developmental processes, such as germination, stomatal development, flowering and senescence, as evidenced by its modification (R. Richter et al., 2013; Zubo et al., 2018). Arabidopsis mutants lacking the GNC and CGA1/GNL genes exhibit a decrease in chloroplast size and a reduction in chlorophyll content of approximately 10-15% compared to their wild-type counterparts (Cackett et al., 2022). The pronounced phenotypic defects in the double mutants suggest that the two transcription factors have overlapping functions in controlling chlorophyll synthesis and chloroplast formation (R. Richter et al.,

2010). Overexpression of GNC or CGA1/GNL in Arabidopsis, as well as CGA1/GNL in rice, accelerates the process of greening during photomorphogenesis, raises chlorophyll levels in mature leaves, and triggers activation of chloroplast development in non-green tissues such as the leaf epidermis and root cells (Richter et al., 2010, 2013; Zubo et al., 2018).

2.6. Convergence of plastid retrograde signaling and photomorphogenesis

During the transition from skotomorphogenesis to photomorphogenesis, plants need to rapidly produce chloroplasts to enable photosynthesis and support autotrophic growth. However, they also need to avoid oxidative damage caused by incompletely assembled photosynthetic machinery. The coordination of plastid-derived retrograde signals and light signals is essential for this process (Pogson et al., 2015). Among the five *GUN* loci that are involved in retrograde signaling, *GUN2 (HY1)* and *GUN3* (*HY2*) have a dual role in both retrograde and light signaling. HY1 and HY2 are responsible for the synthesis of the bilin-type chromophore of phytochrome, the red and far-red light photoreceptor (Bae & Choi, 2008; J. Li et al., 2011). *HY1* and *HY2* were initially identified as components of light signaling pathways (Chory et al., 1989; Parks & Quail, 1991), but the same genes were later found to underlie *gun* mutations as well, indicating their involvement in retrograde signaling (Larkin, 2014; Ruckle et al., 2012; Ruckle & Larkin, 2009, p. 1).

ABSCISIC ACID INSENSITIVE 4 (ABI4) is a nucleus-encoded transcription factor that is involved in mediating downstream transcriptional responses to plastid retrograde signals (Pesaresi & Kim, 2019). It was suggested that ABI4 binds to the CCAC cis-element. This cis-element is often found near another cis-element called the G-box (ACGT), which is the binding site for other transcription factors that regulate gene expression in response to light, such as ELONGATED HYPOCOTYL 5 (HY5). This model proposes that ABI4 competes with these light-regulated transcription factors for binding to the G-box. Consequently, ABI4 can suppress the expression of PhANGs, which are typically induced by light (Koussevitzky et al., 2007; Lee et al., 2007). However, subsequent studies showed that the *abi4* mutant does not show a *gun* phenotype (Gray et al., 2003; Kacprzak et al., 2019). A second genetic screen identified *CRYPTOCHROME 1* (*CRY1*) mutations as *gun* mutants, suggesting that loss of cryptochrome function represses *PhANG* expression by altering the regulatory role of HY5 (Ruckle et al., 2007; Ruckle & Larkin, 2009, p. 1).

Previous studies have reported that *gun1* mutants have impaired chloroplast development when exposed to light after being grown in the dark (Mochizuki et al., 1996; Wu, Meyer, Wu, et al.,

2019 a). This implies that plastid signals and light signals interact during the transition from skoto- to photomorphogenesis. A model was suggested to explain this interaction, involving GUN1, PHD-TYPE TRANSCRIPTION FACTOR WITH TRANSMEMBRANE DOMAINS (PTM) and ABI4 as regulators of light signaling mediated by CONSTITUTIVE PHOTOMORPHOGENIC1 (COP1) and HY5 (Xu et al., 2016). According to this model, a retrograde signal is transmitted by the GUN1 in conjunction with the PTM and ABI4. This signal subsequently modulates the light signal that is transduced by COP1 and HY5. However, this model has been challenged by other experiments that show that ptm and abi4 mutants do not exhibit the gun phenotype (Kacprzak et al., 2019; Page et al., 2017). An alternative hypothesis is that gun1 mutants have reduced protein import into plastids, which affects chloroplast biogenesis (Wu, Meyer, Richter, et al., 2019b).

The transcriptional regulator GLK1 plays a crucial role in photomorphogenesis and serves as a significant mediator of retrograde signals from plastids (Kakizaki et al., 2009). The PHYTOCHROME-INTERACTING FACTOR 4 (PIF4), a key repressor of photomorphogenesis, directly targets the GLK1 gene (Oh et al., 2012). During photomorphogenesis, phytochrome-mediated degradation of PHYTOCHROME-INTERACTING FACTORs (PIFs) leads to increased GLK1 expression and upregulation of photosynthesis-related genes (Martín et al., 2016). However, under stress or when plastids are dysfunctional, GLK1 expression is repressed through a GUN1-dependent mechanism, counteracting the phytochrome signal and slowing down photomorphogenesis (Martín et al., 2016).

The perception of red and far-red light involves retrograde signaling mechanisms (Wu & Bock, 2021). One of them is the regulation of the nuclear localization of phytochrome B (phyB). phyB transport to the nucleus to activate light-responsive genes is facilitated by binding to HEMERA (HMR), a protein that is a part of the PEP complex. HMR also interacts with other PEP-related proteins and regulates plastid gene expression (Nevarez et al., 2017). HMR and other PEP-related proteins, such as REGULATOR OF CHLOROPLAST BIOGENESIS (RCB), NUCLEAR CONTROL OF PEP ACTIVITY (NCP), and PAP8, are found in both plastids and the nucleus, suggesting a connection between the plastid genome expression pathway and phyB-mediated light signaling (Liebers et al., 2020; Nevarez et al., 2017). Another example of dual localization is the transcription factor HvCMF7 in barley and its Arabidopsis homolog CIA2, which affect chloroplast development, possibly by regulating

plastid ribosome formation (Gawroński et al., 2021; M. Li et al., 2019). These findings indicate that many transcriptional regulators may act as key mediators of intercellular communication by being present in both chloroplasts and the nucleus (Wu & Bock, 2021).

In summary, the convergence of retrograde plastid signaling with light signaling during photomorphogenesis is intricately demonstrated through the dual roles of GUN2 (HY1) and GUN3 (HY2). These loci are instrumental in the biosynthesis of the chromophore for phytochrome and are also integral to retrograde signaling. These components highlight the complex interplay between different signaling pathways. The involvement of ABI4, CRY1, and HY5 further emphasises this complexity. Despite previous hypotheses about ABI4's role in retrograde signaling, new studies bring these assumptions into question, illustrating the dynamic and evolving understanding in this field. The interaction between CRY1 and HY5, particularly in regulating PhANG expression, underscores the nuanced regulatory mechanisms at play. Traditional accepted models involving GUN1, PTM, and their regulatory influence through COP1 and HY5 are being reevaluated in light of emerging experimental data. This intricate scenario is further complicated by the discovery of transcription factors such as HvCMF7 / CIA2, which exhibit dual localisation, indicating their multifunctional roles in both chloroplast and nuclear regulatory processes, and highlighting the complex network of interactions that govern plant photomorphogenesis.

2.7. Epigenetic regulation of gene transcription

At the heart of nuclear gene transcription, the role of epigenetic mechanisms such as DNA methylation and histone modification is of paramount importance (Hemenway & Gehring, 2023). Central to these mechanisms is the structure and modification of chromatin, which plays a crucial role in the regulation of gene expression. Chromatin, primarily composed of DNA wound around histone proteins, forms the structural basis of the eukaryotic genome. The basic unit of chromatin is the nucleosome, which consists of a segment of DNA wrapped around an octamer of histone proteins. This octamer is typically made up of pairs of four core histone proteins: H2A, H2B, H3, and H4 (Samo et al., 2021). Each histone protein is characterized by a structured C-terminal core and an unstructured N-terminal tail domain. The N-terminal tails, known for their flexibility, are often subject to a variety of posttranslational modifications. These include acetylation, methylation, ubiquitination of lysine residues, citrullination of arginine residues, phosphorylation of serine, threonine, and tyrosine residues (Yamaguchi, 2021).

Embedded in the complexity of chromatin structure is the histone H1, often referred to as the linker histone. H1 lies outside the nucleosome core, binding the DNA between nucleosomes and helping to compact the chromatin into higher-order structures (Fyodorov et al., 2018). This compaction is vital for the dense packing of DNA in the nucleus and has significant implications for gene regulation. The presence and post-translational modifications of histone H1, along with core histones, are key in controlling the accessibility of transcriptional machinery to the underlying DNA (Rutowicz et al., 2019)

Histone modifications play a pivotal role in this regulation, coordinated by a diverse group of enzymes known as 'writers', such as histone methyltransferases (HMTs) or histone acetyltransferases (HATs). HMTs modify specific residues on histones, affecting gene expression patterns, while HATs facilitate transcription by loosening chromatin structure (Fang et al., 2023). Conversely, 'erasers' like histone demethylases, including JUMONJI DOMAIN-CONTANING PROTEIN (JMJ) domain-containing proteins, and histone deacetylases (HDACs), reverse these modifications, highlighting the dynamic nature of chromatin regulation (Morera et al., 2016). Complementing these are 'readers' - proteins that recognize specific histone modifications and translate them into biological outcomes. Chromodomains and bromodomains, which bind to methylated and acetylated lysine residues respectively, are prime examples of such 'readers', thus influencing gene expression patterns (Jain et al., 2020).

Chromatin remodelers such as BRAHMA (BRM) and PICKLE (PKL) are crucial in repositioning and restructuring nucleosomes, thereby modulating the access of transcription machinery to DNA. These remodelers, while not altering histone modifications directly, affect the overall architecture of chromatin, playing a vital role in gene regulation (Liu et al., 2022). They are part of a complex network of epigenetic regulation, working synergistically with the 'writers', 'erasers', and 'readers' to ensure precise control over gene expression.

The intricate organization of chromatin and its modifications constitute a fundamental layer of epigenetic regulation. This regulation is a dynamic process, influencing gene expression patterns without altering the underlying DNA sequence, thereby playing a pivotal role in cellular differentiation, development, and response to environmental cues (Berger et al., 2009).

2.7.1. Polycomb Repressive Complexes (PRC): PRC2 and PRC1

Polycomb Group (PcG) proteins play a crucial role in epigenetic regulation by maintaining developmental gene repression (Baile et al., 2022). These proteins are organized into two

distinct and important multi-protein complexes known as Polycomb Repressive Complex 1 (PRC1) and Polycomb Repressive Complex 2 (PRC2) (Margueron & Reinberg, 2011), which are responsible for establishing and preserving transcriptional repression by catalysing repressive histone post-translational modifications.

PRC2, found in various multicellular organisms, catalyzes the trimethylation of histone H3 at lysine 27 (H3K27me3), an essential epigenetic mark associated with gene repression (Mozgova et al., 2015). Loss of core PRC2 subunits in plants and animals leads to developmental defects due to the disruption of H3K27me3 levels in PRC2 target genes (Deevy & Bracken, 2019; Mozgova et al., 2015).

While the subunits of PRC2 are conserved across multicellular organisms, the number of genes encoding each subunit varies among species (Mozgova et al., 2015). For instance, Drosophila PRC2 comprises four main components: Enhancer of zeste (E(z)), Suppressor of zeste 12 (Su(z)12), Extra sex comb (Esc), and p55 (Margueron & Reinberg, 2011). In humans, two copies of E(z)—EZH1 and EZH2—exist (Ciferri et al., 2012). Arabidopsis possesses three homologous genes for the E(z) subunit: CURLY LEAF (CLF), SWINGER (SWN), and MEDEA (MEA); three for Su(z)12: EMBRYONIC FLOWER 2 (EMF2), VERNALIZATION 2 (VRN2), and FERTILIZATION INDEPENDENT SEED 2 (FIS2); one gene for Esc: FERTILIZATION INDEPENDENT ENDOSPERM (FIE); and five genes for p55: MULTICOPY SUPRESSOR OF IRA 1-5 (MSI1-5) (Hennig et al., 2005; Kenzior & Folk, 1998).

In Arabidopsis, CURLY LEAF (CLF) and SWINGER (SWN) function as catalytic subunits of the PRC2 complex in the sporophyte. The third catalytic subunit MEDEA (MEA), contributes to H3K27me3 deposition but is predominantly expressed during female gametophyte and early seed development (Chanvivattana et al., 2004; Shu et al., 2019, simonini et al 2021). In particular, MEA's functions are distinct from CLF and SWN, which are primarily active during vegetative growth and inflorescence development (Y. Liu et al., 2022). FERTILIZATION INDEPENDENT SEED2 (FIS2)-associated PRC2 has a pivotal role in seed development (Köhler et al., 2003). The VERNALIZATION2 (VRN2)-PRC2 complex contributes to vernalization-triggered flowering through its regulation of *FLOWERING LOCUS C* (*FLC*) (De Lucia et al., 2008). Furthermore, EMBRYONIC FLOWER2 (EMF2)-PRC2 is indispensable for sustaining the vegetative phase by suppressing the developmental MADS box genes including *AGAMOUS* (*AG*), *APETALA3* (*AP3*) or *PISTILLATA* (*PI*), and its disruption leads

to various aberrant phenotypes such as early flowering, leaf curling, terminal flowers, and anomalous floral organs, attributed to widespread misregulation of multiple flower homeotic genes (Pu et al., 2013).

Previous research has demonstrated the significant roles of PRC2 proteins in numerous growth and developmental processes (C. Li et al., 2015; Shu et al., 2019). For example, CLF has been linked to the regulation of leaf and flower morphology by repressing *AGAMOUS* (*AG*) and *SHOOT MERISTEMLESS* (*STM*) (Schubert et al., 2006). Additionally, CLF is involved in the epigenetic repression of the major flowering repressor *FLOWERING LOCUS C* (*FLC*), whose repression is triggered by vernalization (Heo and Sung, 2011).

PRC2 lacks DNA-binding domain and among other mechanisms mainly relies on interaction with various transcription factors that interact with PRC2 subunits to recruit the complex to DNA target genes in a sequence-specific manner (Xiao et al., 2017). For instance, CLF and SWN are recruited to target genes by TELOMERE-REPEAT-BINDING FACTORS (TRBs) through telobox-related motifs (Zhou et al., 2018). In addition, VIVIPAROUS1/ABI3-LIKE1 (VAL1) and VAL2 are transcription factors required for PRC2 recruitment during the seed-to-seedling transition (N. Chen et al., 2018; H. Jia et al., 2013; Yuan et al., 2020).

Polycomb repressive complex 1 (PRC1) mediates histone H2A ubiquitination (H2Aub) (Beisel & Paro, 2011; Simon & Kingston, 2009). Alone or in combination with H3K27me3, deposition of H2Aub compacts chromatin and restricts gene accessibility, particularly crucial during development (Francis et al., 2004). The composition of plant PRC1 complexes is not well determined yet. Nevertheless, the catalytic components appear to be RING domain proteins (RING1, BMI1). TERMINAL FLOWER 2/LIKE HETEROCHROMATIN PROTEIN 1 (TFL2/LHP1), EMBRYONIC FLOWER 1 (EMF1), and VERNALIZATION 1 (VRN1) have been proposed to also comprise the PRC1 (Calonje, 2014). In Arabidopsis, RING1-like (AtRING1A, AtRING1B) and BMI1-like proteins (AtBMI1A, AtBMI1B, AtBMI1C) collaborate with PRC2 for post-embryonic repression, atalysing H2A monoubiquitination (Bratzel et al., 2010). LHP1/TFL2, a versatile PcG protein, interacts with diverse partners to reach chromatin targets, impacting leaf morphogenesis, flowering, and stress responses in loss-of-function mutants (Parihar et al., 2019). EMF1, lacking conserved motifs, interacts with RING1A-B and BMI1A-C, is crucial for H2Aub, and mutations lead to developmental transition within the growth meristems. VRN1, involved in vernalization, could be another

PRC1 subunit, but its function and interactions remain unknown (Berke & Snel, 2015; Pu et al., 2013).

Overall, PRCs have been mainly associated with developmental gene repression. Recently emerging reports indicate involvement of PRCs in the regulation of environmental and defence response genes (Ramirez-Prado et al., 2019). There is very little evidence and understanding of PRCs function during repression of genes whose transcriptional level dynamically responds to environmental cues, including light availability.

2.7.2. Chromatin modifiers associated with Polycomb and/or light signalling

PcG repression is counteracted by Trithorax-group (TrxG) protein complexes that serve as activators of PcG-repressed genes (Kuroda et al., 2020; Schuettengruber et al., 2017). This group of protein complexes includes activating histone methyltransferases (depositing e.g. H3K3me3, H3K36me3 etc.), histone acetyltransferases and multiple ATP-dependent nucleosome remodellers, which use ATP hydrolysis to alter, displace or reposition nucleosomes and create an open chromatin state that allows the access of transcriptional factors or other regulators (Clapier & Cairns, 2009; Saha et al., 2006).

Interplays between PcG and other TrxG histone methyltransferases have been explored. For example, CLF physically interacts with ARABIDOPSIS HOMOLOG OF TRITHORAX 1 (ATX1), a histone methyltransferase catalyzing H3K4me3, to regulate the *AG* silent state in young seedlings (Saleh et al., 2007; Shu et al., 2020). CLF also collaborates with SET DOMAIN GROUP 8 (SDG8) (Bian et al., 2016; Tang et al., 2012), a histone methyltransferase (HMT) that targets histone H3 lysine 36 (H3K36) in plants (X. Wang et al., 2014). SDG8 catalyzes the di- and trimethylation (H3K36me2 and H3K36me3) and inhibits the monomethylation (H3K36me1) of this residue, thereby altering the chromatin structure and gene expression (Guo et al., 2010). SDG8 plays a crucial role in regulating various biological processes in plants, such as flowering time, shoot branching, reproductive development, carotenoid metabolism, seed gene expression, fungal defense, and innate immunity (Y. Li et al., 2015). BRAHMA (BRM) and SPLAYED (SYD) are two SWItch/Sucrose Non-Fermentable (SWI/SNF)-type ATPases of the TrxG. BRM and SYD counteract CLF activity function at *AGAMOUS* (*AG*) and *APETALA 3* (*AP3*) during flower development (Shu et al., 2020). BRM restricts CLF/SWN occupancy and activity at the SHORT VEGETATIVE PHASE (SVP) gene during vegetative development (C. Li et al., 2015, 2016; Shu et al., 2020). Recent evidence also suggests that SYD, similar to BRM, exhibits both antagonistic and

collaborative roles with PRC2 in mediating H3K27me3 deposition in Arabidopsis seedlings (Shu et al., 2021). The chromatin SWI/SNF ATP-dependent (CHD)-type remodeller protein PICKLE (PKL) is required for root meristem growth, acting antagonistically to CLF in the determination of root meristem activity (Aichinger et al., 2011). Recent findings indicate that PKL and CLF also function together at a common set of genes and are both necessary for maintaining genome-wide H3K27me3 levels (Aichinger et al., 2011; Shu et al., 2020).

H3K27me3 deposition by PRC2 is counteracted by catalytic complexes that mediate removal (demethylation) of the repressive marks (Crevillén, 2020). Among these are the JMJ-domain demethylases such as EARLY FLOWERING 6 (ELF6), RELATIVE OF ELF6 (REF6), JMJ13, and JMJ30 (Crevillén et al., 2014; He et al., 2021; Lu et al., 2011; Zheng et al., 2019). Antagonistic interactions are frequently facilitated by competitive binding at identical chromatin sites (Zhu et al., 2020). However, PRC2 shows a preference for distinct motifs, such as the telobox and GAGA motifs, indicating that competitive antagonism between JMJ proteins (specifically REF6) and PRC2 is improbable as they do not share overlapping binding patterns (Yamaguchi, 2021). Notably, REF6 is found at the periphery of H3K27me3 regions, which are occupied by PRC2 (Yan et al., 2018). The spread of H3K27me3 observed in *ref6 elf6 jmj13* triple mutants implies that REF6 binding acts to inhibit the extension of H3K27me3. However, the specific mechanisms through which ELF6 and JMJ13 contribute to preventing this spreading are yet to be fully understood (Yamaguchi, 2021).

PRC2 repression and H3K27me3 deposition has been connected to the presence of the linker histone H1. Plants carrying mutations in the three *H1* genes present in *Arabidopsis* display some developmental phenotypes resembling PRC2 depletion mutants (Rutowicz et al. 2019, Celia Baroux and Kinga Rutowicz, University of Zurich, personal communication).

Histone acetylation and deacetylation are reversible modifications that affect gene regulation by altering the chromatin structure. The balance between histone acetylation and deacetylation is maintained by the antagonistic activities of histone acetyltransferases (HAT) and deacetylases (HDAC), which catalyze the addition or removal of acetyl groups to lysine residues on the N-terminal tails of histone H3 and H4 (Pandey et al., 2002). In *Arabidopsis thaliana*, several lysine sites on histone H3 (K9, K14, K18, K23, and K27) and H4 (K5, K8, K12, K16, and K20) have been reported as targets of acetylation and deacetylation (Benhamed et al., 2006; Servet et al., 2010). GENERAL CONTROL NON-REPRESSIBLE 5 (GCN5) or HISTONE ACETYLTRANSFERASE OF THE GENERAL CONTROL NON-

REPRESSIBLE 5 (HAG1) (AtGCN5/HAG1) is a key HAT that modulates gene expression in various plant developmental pathways and environmental responses (Servet et al., 2010). Mutations in AtGCN5/HAG1 cause several phenotypic defects, such as reduced plant height, impaired meristem activity, abnormal root and leaf morphology, defective floral organ specification, and diminished responsiveness to light and cold stimuli (Bertrand et al., 2003; Kornet & Scheres, 2009; Servet et al., 2010; Long et al., 2006; Vlachonasios et al., 2003). Moreover, GCN5, HD1, and TAF1/HAF2 interact to regulate histone acetylation required for light-responsive gene expression (Benhamed et al., 2006). These findings suggest that AtGCN5/HAG1 plays a role in diverse developmental and environmental processes in Arabidopsis.

There are 12 RPD3-like HDACs, among which HDA5, HDA6, HDA9, and HDA19 have been extensively studied (Peng et al., 2017) in *Arabidopsis thaliana*. HDA6 plays a key role in DNA methylation, heterochromatin formation, and various signaling pathways involving phytohormones, circadian rhythms, stress responses, and flowering time (Ning et al., 2019). HDA19 also participates in many developmental processes, such as embryogenesis, germination, floral development, phytohormone signaling, and stress responses, either redundantly or independently of HDA6 (L.-T. Chen & Wu, 2010; Krogan et al., 2012). According to (Ning et al., 2019), the HDA19 histone deacetylase complex also plays a role in regulating the timing of flowering in a manner that is dependent on photoperiod.

Altogether, a multitude of chromatin-modifying proteins complexes are known to act synergistically or antagonistically to PRCs and to also regulate environmental responses. It however remains unknown whether and how these mechanisms contribute to the regulation of photosynthesis-associated nuclear gene (PhANG) expression This thesis extends previous preliminary results of the lab that identified *gun* phenotype in PRC2 mutants. The aim of this work is to screen for the presence of *gun* phenotype in mutants affected in light and/or PRC2-related pathways to determine the extent of their involvement in PhANG regulation.

3. Material and Methods

3.1 Chemicals

GeneRuler 1kb RNA ladder (#SM0311, Thermo Fisher Scientific); DNA Gel Loading Dye 6x (#R0611,Thermo Scientific; #B7024S, BioLabs); Ethanol 96% (#70390, Penta); Murashige & Skoog medium including vitamins (= MS) (#M0255, Duchefa); sucrose; LB Broth with agar (#L2897, Sigma Aldrich); Dithiothreitol (DTT); Lincomycin; DNase I (lyophilized, Thermo Fisher Scientific); RNA Binding Beads (Thermo Fisher Scientific); DNase I Reconstitution Buffer (Thermo Fisher Scientific); 2X DNase I Buffer (Thermo Fisher Scientific); Lysis Buffer (Thermo Fisher Scientific); Wash Solution 1 Concentrate (Thermo Fisher Scientific); Wash Solution 2 Concentrate (Thermo Fisher Scientific); Manganese Chloride Solution (Thermo Fisher Scientific); RNA Rebinding Buffer (Thermo Fisher Scientific); Water,Nuclease-free (Thermo Fisher Scientific); RevertAid RT (200 U/µL, Thermo Scientific, #K1622), RiboLock RNase Inhibitor (20 U/µL, Thermo Fisher Scientific, #K1622), 5X Reaction Buffer (250 mM Tris-HCl (pH 8.3), 250 mM KCl, 20 mM MgCl2, 50 mM DTT; Thermo Fisher Scientific), 10 mM dNTP Mix, $Oligo(dT)_{18}$ Primer, 100 µM (Thermo Fisher Scientific), Random Hexamer Primer, 100 µM, (Thermo Fisher Scientific), Forward GAPDH Primer, 10 µM (Thermo Fisher Scientific), Reverse GAPDH Primer, 10 µM (Thermo Fisher Scientific), Control GAPDH RNA, 0.05 µg/µL (Thermo Fisher Scientific); Dentist machine (ivoclar vivadent, SILAMAT)

3.2 Kits

The listed buffers in 2.1 are included in the following kits:

- MagMAX Plant RNA Isolation Kit (A33784, Thermo Fisher Scientific)
- Thermo Scientific RevertAid First Strand cDNA Synthesis Kit (#K1622)

3.3 Prepared buffers and media

To prepare RNA wash solutions from concentrates, 110 mL of 96% ethanol was added to each bottle of wash solution 1 concentrate (MagMAX Plant RNA Isolation Kit), mixed and stored at room temperature. Subsequently, 96% ethanol was added to each bottle of Wash Solution 2 Concentrate (MagMAX Plant RNA Isolation Kit), mixed and stored at room temperature.

DNase I solution: 440 µL of DNase I Reconstitution Buffer was added to each vial of DNase I (lyophilized). Followed by an incubation for 5 minutes and stored in aliquots at –20°C.

3.4 Devices

GFPCam FC 800-C/1010GFP (Photon Systems Instruments, PSI, Czech Republic); Laminar hood (SCS 1-5, MERCI; HB2448, Holten LaminAir; OSN-5, MERCI); Thermo-Shaker (TS 100C, BioSan); microwave (SAMSUNG); electrophoresis tanks (Cleaver Scientist); electrophoresis power supply (Power Pac Basic, BIO RAD); ChemidDoc™MP Imaging System (BIO RAD); Spectrophotometer (NanoDrop™ ND-1000, Thermo Scientific); minicentrifuge (M-6, BOECO), Eppendorf tubes (1.5 ml; 2 ml); tweezers; stratification box; Petri plates; glass beads; Pipettes [(2 µl, 10 µl, 20 µl, 200 µl, 1000 µl), Eppendorf Research Plus; 10 µl, DISCOVERY Comfort]; centrifuge (Z 216 MK, HERMLE; UNIVERSAL 320 R, Hettich ZENTRIFUGEN); qPCR cyler (CFX Connect Real-Time PCR Detection System #1855201)

3.5 Plant material

Arabidopsis thaliana, ecotype Colombia (Col-0) was used as control. *gun1-1* mutant plants were used as positive controls indicating *gun* phenotype. Additionally, mutant plants carrying mutations in various chromatin-related proteins as specified in Table 1 were analysed. A preliminary study was conducted to validate the assay's efficacy. This study was confined to mutants in set 1. The assay focused on four photosynthetic-associated nuclear genes: *CHLH*, *GUN4*, *PHOTOSYSTEM I LIGHT HARVESTING COMPLEX GENE 1* (*LHCA1*), *LIGHT HARVESTING COMPLEX OF PHOTOSYSTEM II 5 (LHCB5),* and *Glutamyl-tRNA reductase 1 (HEMA1*). Based on the initial findings, the optimized assay was used to analyse all mutants, separated into three experimental groups (set 2 to set 4: Table 2) that were tested in three independent experiments to enable the processing of the samples.

Table 1: Arabidopsis thaliana mutants with references employed in the Study

Gene affected	**Gene ID (TAIR)**	**Alelle name**	**Public database ID**

GUN1	*AT2G31400*	*gun1-1*	Susek et al., 1993, Cell: donated by Dr. Terry
CLF	*AT2G23380*	*clf-29*	SALK_021003
SWN	*AT4G02020*	*swn-3*	SALK_050195
FIE	*AT3G20740*	*fie/+*	GABI KAT
MSI1	*AT5G58230*	*msi1-1 (co-suppresion) - msi1 cs*	Hennig et al 2003
EMF2	*AT5G51230*	*emf2*	SALK_011550C
VRN2	*AT4G16845*	*vrn2*	SALK_201153
LHP1	*AT5G17690*	*lhp1-6*	SALK_011762
PKL PKR2	*AT2G25170, AT4G31900*	*pkl-1 pkr2*	*pkr2-1*:SALK 109423 and *pkr2-2*: SALK 115303
HSI2/VAL1	*AT2G30470*	*val1-2*	SALK_088606C
HSL1/VAL2	*AT4G32010*	*val2-3*	SALK_059568C
H1.1, H1.2, H1.3	*AT1G06760 (H1.1), AT2G30620 (H1.2)*	*3h1 (h1.1 h1.2 h1.3)*	Dr. Rutowicz, Dr. Baroux
H1.3	*AT2G18050 (H1.3)*	*h1.3*	Dr. Rutowicz, Dr. Baroux
H1.1, H1.2	*AT1G06760 (H1.1), AT2G30620 (H1.2)*	*h1.1 h1.2*	Dr. Rutowicz, Dr. Baroux
SWN H1.1, H1.2, H1.3	*AT4G02020, AT1G06760 (H1.1), AT2G30620 (H1.2), AT2G18050 (H1.3)*	*swn-7 3h1*	Dr. Rutowicz, Dr. Baroux
CLF H1.1, H1.2	*AT2G23380, AT1G06760 (H1.1), AT2G30620 (H1.2)*	*clf-29 h1.1 h1.2*	Dr. Rutowicz, Dr. Baroux
CLF H1.1, H1.2, H1.3	*AT2G23380, AT1G06760 (H1.1), AT2G30620 (H1.2), AT2G18050 (H1.3)*	*clf-29 h1.1 h1.2 h1.3*	Dr. Rutowicz, Dr. Baroux
BRM	*AT2G46020*	*brm-1*	SALK_030046
SDG8	*AT1G77300*	*sdg8-IM2*	SALK_026442
JMJ20	*AT5G63080*	*jmj20-IM1*	SALK_202511C

GCN5	*AT3G54610*	*gcn5-IM1*	SALK_150784
TAF1=HAF1	*AT1G32750*	*taf1-3=haf1-3*	SALK_110848
HDA6	*AT5G63110*	*axe1-5 , hda6-6*	CS66153
HDA6	*AT5G63110*	*had6-IM1*	SALK_201895C
HDA19 = HD1	*AT4G38130*	*hda19-3*	SALK_139445
GUN2	*AT2G15360*	*gun1-2*	Susek et al., 1993, Cell: donated by Dr. Terry
CLF	*AT2G7340*	*clf-30*	SALK_079387

The experiment was conducted once for each set of mutant plants, where each mutant was analysed in three technical replicates. However, due to the extensive array of mutants and the time constraints imposed by the thesis timeline, biological replicates were not incorporated. Therefore, statistical analysis was not performed.

Table 2: Experimental setup for the gun phenotype assessment

Set 1	**Set 2**	**Set 3**	**Set 4**
col-0	*col-0*	*col-0*	*col-0*
gun1-1	*gun1-1*	*gun1-1*	*gun1-1*
clf-29	*swn-3*	*brm1*	*3h1*
swn-3	*swn-3-/gun1*	*sdg8*	*h1.3*
pkl pkr2	*emf2*	*gcn5/hat1*	*h1.1 h1.2*
ring1a bmi1a	*vrn2*	*hda6-6*	*swn-7 3h1*
ring1a bmi1c	*lhp1*	*taf1-3*	*clf-29 h1.1 h1.2*
	val1-2	*hda6-1*	*clf-29 h1.1 h1.2 h1.3*
	val2-3	*hda19-3*	
	fie/+		
	msi1-1		

3.6 Surface sterilization of seeds

The germination of *Arabidopsis* seeds on Petri plates containing artificial growth medium requires a sterile environment to avoid or reduce the risk of microbial contamination. In this study, ethanol sterilization was used due to its simplicity and efficacy. The seeds were immersed in 1 ml of 70% ethanol for a duration of 11 minutes. Subsequently, the ethanol solution was substituted with 1 ml of 90% ethanol for an additional 3 minutes. The seeds were subjected to a drying process inside the open microtube, placed in laminar flow box, for a period of 50 minutes. This procedure resulted in minimal contamination and a high germination rate of Arabidopsis seeds.

3.7 Seed sowing on Petri plates containing mineral growth medium

Each set (genotype) of mutant seeds was cultivated on three plates without and three plates with lincomycin (0.5 mM) in the growth medium. The seeds were stratified in a dark cold room at 4 °C for three days to synchronize germination. After stratification, the seeds were transferred to growth chambers (Photon Systems Instruments) and grown under medium light (125 µmol.m-2.s-1) for seven days. To avoid the confounding effects of different sowing times, all genotypes in the same set were germinated simultaneously.

3.8 Chlorophyll fluorescence

Photosynthetic-related parameters were determined analysing seedlings grown for 7 days. A closed GFPCam FC 800-C/1010GFP (Photon Systems Instruments, PSI, Czech Republic) was used to determine the various parameters based on chlorophyll fluorescence. Seedlings were acclimated to darkness for 20 min. The minimal fluorescence level (F_0) was measured by estimating the modulated light (0.1 µmol m^{-2} s^{-1}). The intensities of the actinic light and saturating light settings were 280 µmol mol−2 s−1 and 2500 µmol mol−2 s−1 PAR, respectively. The maximum quantum yield of PSII (Fv/Fm) were measured. The Fv/Fm ratio, a key parameter obtained through chlorophyll fluorescence analysis, reflects the efficiency of light energy utilization for photosynthesis and the parameter decreases under stress conditions, indicating photoinhibition and reduced PSII activity. Consequently, fluorescence analysis can serve as a reliable measure of the effectiveness of lincomycin treatment. Chlorophyll fluorescence was exclusively conducted on the seedlings in Set 1, as the efficacy of lincomycin can be extrapolated from the phenotypic observation of the seedlings at seven days old (Figure 3).

3.9 Total RNA isolation

The shoots of the seedlings in the preliminary study, as well as those from Sets 1 through 4, were collected after 7 in the growth chambers. Approximately 50 mg of shoot tissue was harvested from each plant at 10-11 am (Zeitgeber Time ZT 4-5) and placed in an autoclaved microtube containing glass beads. The tubes containing plant material were immediately frozen in liquid nitrogen. Plant material was homogenised. Thermo Scientific MagMAX™ Plant RNA Isolation Kit was used to isolate total RNA from the plant tissue according to the manufacturer's instructions.

3.9.1. Disruption of plant tissue

The plant tissue, approximately 50 mg, was homogenized with a bead beater (ivoclar vivadent, SILAMAT) in the presence of glass beads for 20 seconds until it was completely pulverized. Next, 600 µL of lysis buffer (pH 8.0) containing 12 µL of 2 M DTT was added to each tube and briefly vortexed to mix. The samples were then incubated at 56 °C for 5 minutes and centrifuged for 10 minutes at maximum speed (20,000 × g) to remove the plant lysates. 400 µL of the supernatant was then transferred to a new 1.5 ml microcentrifuge tube for manual purification.

4.9.2. RNA Purification

RNA was isolated from plant lysates using RNA-binding beads as follows. First, 25 µL of RNA binding beads were added to 400 µL of plant lysate in a microcentrifuge tube. Next, 400 µL of 96-100% ethanol was added to the mixture and vortexed for 10 seconds at a high setting to ensure homogeneity. The samples were then briefly centrifuged to collect the liquid at the bottom. After that, the samples were placed on a magnetic stand for 2 minutes until the solution cleared and the beads were collected on the magnet. The supernatant was then carefully discarded without removing the tubes from the magnetic stand. Finally, to wash the RNA in the beads, 700 µL of washing solution 1 (Table 3) was added to the samples and vortexed for 10 seconds at high setting.

Table 3: *Wash Solutions*

	Wash Solution 1	**Wash Solution 2**
Concentrated buffer	110 mL	50 mL
Ethanol (96-100%)	**110 mL**	**200 mL**

Total volume:	220 mL	250 mL

3.9.3. DNase I treatment

Any contaminating genomic DNA was removed from the RNA-bound beads by adding 200 µL of DNase I Master Mix (Table 4) to each sample and mixing well by pipetting. The samples were then incubated in a thermomixer at 37 ° C and 350 rpm for 15 minutes to allow the DNase I enzyme to digest the DNA. After incubation, 150 µL of rebinding buffer was added to each sample to restore the optimal binding conditions for RNA. Next, 400 µL of 96-100% ethanol was added to each sample to precipitate the RNA on the beads. The samples were then washed with 700 µL of wash solution 1 and twice with wash solution 2, following the same procedure as before. Finally, the samples were dried on the magnetic stand with the lid open for 5 minutes to evaporate any residual ethanol.

Table 4: DNase 1 master mix

Component	Volume per sample
2X DNase I buffer	100 µL
DNase I solution	4 µL
Manganese chloride solution	20 µL
Nuclease-Free Water	76 µL
Total DNase I Master Mix	200 µL

3.9.4. Elution of RNA

The beads were resuspended in 30 µL of nuclease-free water and vortexed at high speed for 10 seconds to ensure thorough mixing. The sample was then placed on a magnetic stand and incubated for 2 minutes until the beads were magnetically separated and the supernatant was clear. The supernatant containing the RNA was carefully transferred to a new tube and stored at -80 ° C until further analysis. The quantity and quality of RNA were assessed by measuring the absorbance at 260 nm and 280 nm using a NanoDrop spectrophotometer (NanoDrop™ ND-1000, Thermo Scientific) and integrity was estimated using 1% agarose gel electrophoresis.

3.10 Agarose gel electrophoresis

A 1% agarose gel was prepared from a mixture of agarose and 1xTris-acetate-EDTA (TAE) buffer. One drop of ethidium bromide was subsequently added for every 50 mL of agarose solution (final concentration 2.5µl EtBr/50 ml buffer). 1 µl of the RNA was mixed with 1 µl of DNA 6x Gel Loading Dye and 4 µl nuclease free water, loaded into the wells of the agarose gel placed in an electrophoresis tank (Cleaver Scientific, n.d.) and separated at 6 V/cm for 40 minutes. The GeneRuler 1kb RNA Ladder was used as an RNA fragment size marker. The result was documented using the bottom UV illumination mode on the ChemiDoc gel documenting system (Bio-Rad).

3.11 cDNA synthesis

cDNA was synthesized using the RevertAid First Strand cDNA Synthesis Kit (Thermo Scientific, #K1622) with 1 µg of total RNA as template. The cDNA first strand synthesis was performed usig a mix of 0.5 µL of random hexamer and 0.5 µL oligo-dT primer to synthesize polyA-containing transcripts as well as transcripts that do not carry polyA, such as organellar transcripts. Nuclease-free water was added to the solution to add up to a total volume of 12 µL. 8 µL of master mix (5) was added to each sample, gently mixed, briefly centrifuged, and incubated for 5 min at 25 °C, followed by 60 min at 42 °C. The reaction was terminated by incubating at 70 °C for 5 min. Finally, the reverse transcription reaction products were diluted 10 times and stored at -20 °C for real-time PCR.

Table 5: Master mix for cDNA synthesis

5X reaction buffer	4 µL
RiboLock RNase Inhibitor (20 U/µL)	1 µL
10 mM dNTP Mix	2 µL
RevertAid M-MuLV RT (200 U/µL	1 µL
Total volume	8 µL

3.12 Real-time PCR

EvaGreenMM-5x (SolisBiodyne, 08-33-0000S) was used as a fluorescent dye-contaning master mix that enabled amplification while monitoring the amount of amplified target in real time using quantitative PCR (qPCR). The qPCR reaction mixture (Table 6) consisted of 2 µL of diluted cDNA template, 4 µL of EvaGreenMM-5x, 13.2 µL of nuclease-free water, and 0.4

µL of each forward and reverse primer specific for each target gene (Table 7). The total reaction volume was 20 µL per sample. A melting curve analysis was performed after the amplification cycle to verify the specificity of the primers and presence of a single amplicon type.

Table 6: PCR-SolisBiodyne (EvaGreenMM-5X)

1 reaction components	20ul reaction
water	13.2
Primer-each	0.4
MM	4
Template	2
Total	20

Table 7: primers for each gene

GENE	Gene ID	Primer ID	F/R	Sequence (5'-3')	Length (nt)	Annealing T(°C)
PP2A (reference)	AT1G13320	AT_001	F	ATTCCGATAGTCGACCAAGC	20	54
		AT_002	R	AACATCAACATCTGGGTCTTCA	22	54
LHCB2.1	AT2G05100	AT_068	F	CTCCGCAAGGTTGGTGTATC	20	55
		AT_069	R	CGGTTAGGTAGGACGGTGTAT	21	55
HEMA1	AT1G58290	AT_076	F	GCTTCTTCTGATTCTGCGTC	20	54
		AT_077	R	GCTGTGTGAATACTAAGTCCAATC	24	54
CHLH (GUN5)	AT5G13630	AT_220	F	AGTGGAGCAACTCTGCATCA	20	56
		AT_221	R	AAAACAGTGATTGCCAGCTTC	21	54
GUN4	AT3G59400	AT_072	F	CAATCTCACTTCGGACCAAC	20	53
		AT_073	R	TTGAAACGGCAGATACGG	18	52
LHCA1	At3G54890	AT_125	F	CCGGGAATGTTGGTCGTAT	19	54
		AT_126	R	GGTCAAACCCAAAGTCACCA	20	55

5. Results

4.1 Seedling growth phenotypes

The initial stage of screening for the gun phenotype in selected mutant seeds involved cultivating the seeds in MS medium. This was performed in controlled growth chambers under a continuous medium light intensity (125 µmol.m^{-2}. s^{-1}) for 7 days.

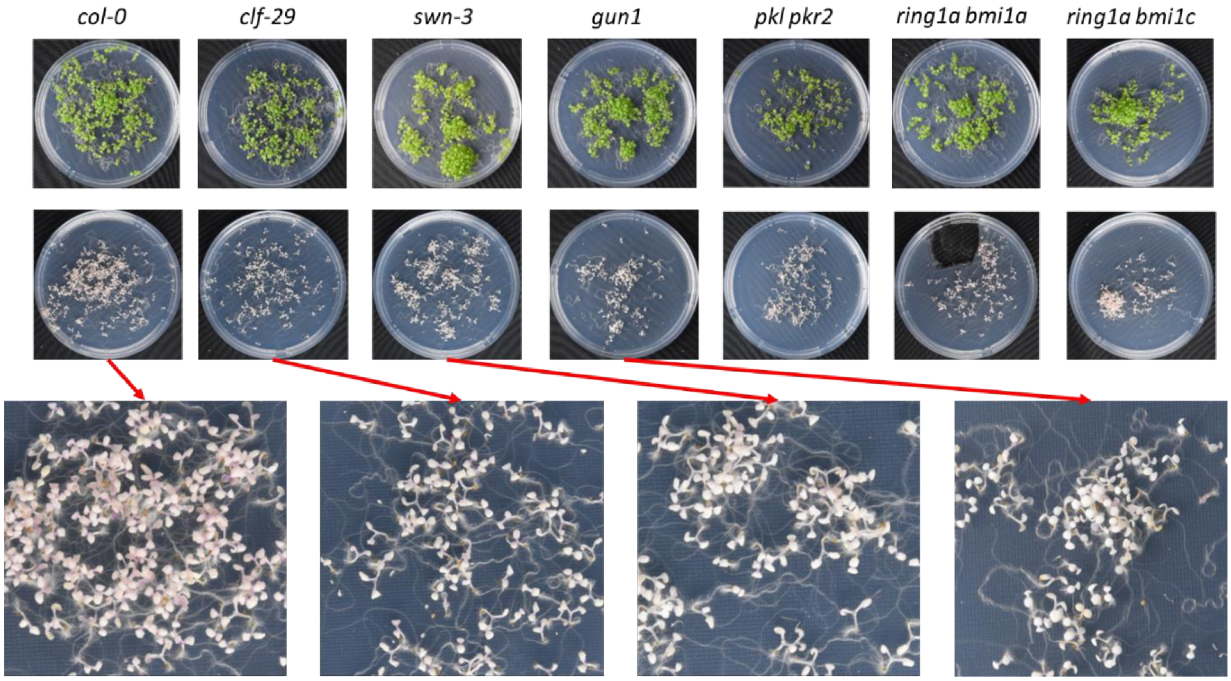

Figure 3: phenotypic comparison of mutants in set 1 of the experimental setup. The seedlings in the first row are the control, while the seedlings in the second row were treated with lincomycin.

Lincomycin treatment has previously been shown to inhibit plastid gene translation and induce retrograde signalling in Arabidopsis (Mulo et al., 2003b). The mutants (Figure 3) were compared to the wild type in terms of plant growth, chlorophyll levels, and cotyledon size. As shown in Figure 3 and in line with expectation, the cotyledons of plant lines exposed to lincomycin did not expand and accumulate chlorophyll (Martín et al., 2016). Chlorophyll fluorescence analysis was performed to assess the effects of lincomycin treatment. The Fv/Fm ratio, a key parameter derived from this technique, was used to evaluate the efficiency of photosystem II (PSII) in converting light energy into chemical energy through photosynthesis (Lichtenthaler et al., 2005). The experiment involved only mutants in set 1. In mature Arabidopsis, an Fv/Fm ratio of 0.81 or higher is considered to mark optimal state in non-stressed mature plants (Zienkiewicz et al., 2015). Plant lines exhibited Fv/Fm ratios between

0.65 and 0.74 ug/ul in control and between 0.095 and 0.313 ug/ul in lincomycin-grown plants (Figure 4).

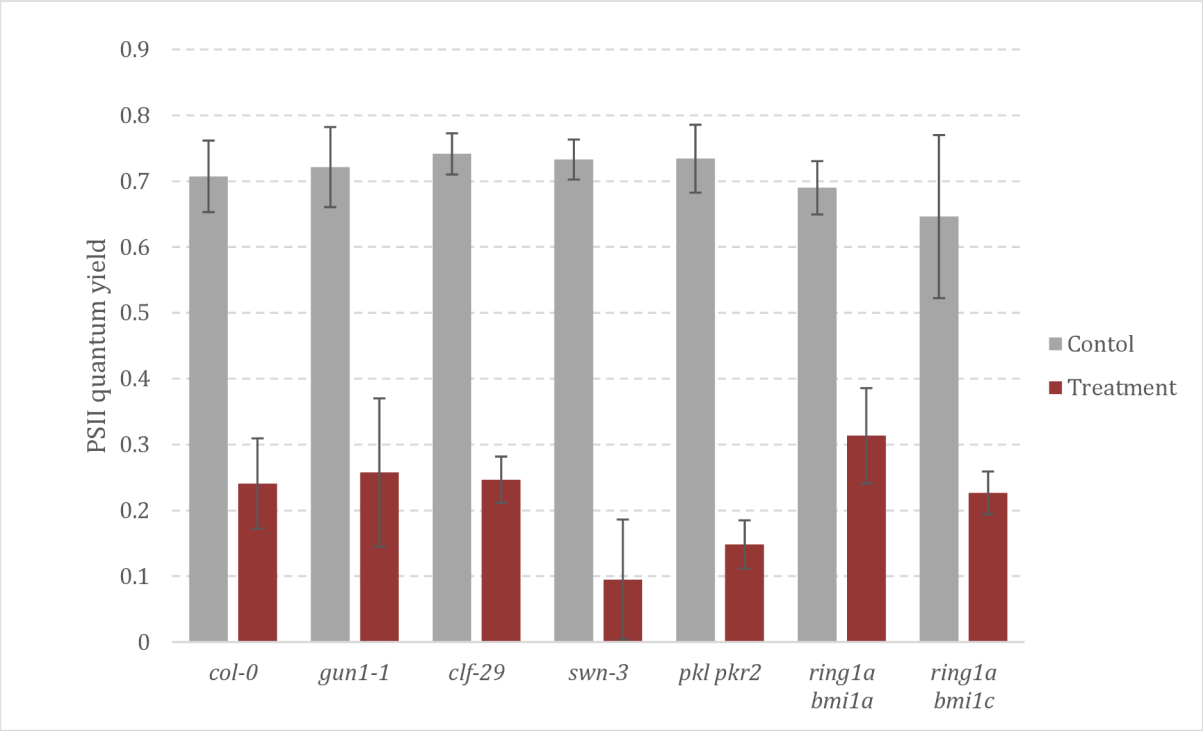

Figure 4: PSII quantum yield in Arabidopsis in set 1 grown under control conditions and in the presence of lincomycin. The analysis was based on three biological replicates and error bars indicate standard deviation.

4.2 Total RNA isolation

RNA isolation using the Thermo Scientific MagMAXTM Plant RNA Isolation Kit led to a successful extraction of total RNA from seedlings . The average RNA yield from 50 mg leaves was between 185.7 and 654.6 ug/ul in the control samples and between 114.7 and 447.4 ug/ul in plants grown in the presence of lincomycin. The purity of isolated RNA was assessed using a NanoDrop spectrophotometer by measuring the absorbance ratio at 260/280 and 260/230 nm (Figure 5).

The integrity of the isolated RNA samples was evaluated by 1% agarose gel electrophoresis. Although the overall RNA bands were distinct indicating intact RNA, some issues were observed. Some of the RNA bands were thicker and not fully separated from the 18S RNA band.

	Plant ID	Name	RNA Conc. (ng/µl)	260/280	260/230
Control	1	*Col-0*	654.6	2.27	1.81
	2	*clf-29*	263.4	2.27	1.65
	3	*swn-3*	416.8	2.21	2.17
	4	*gun1*	503.4	2.26	2.09
	5	*pkl pkr2*	185.7	2.28	1.81
	6	*ring1a bmi1a*	387.1	2.25	2.16
	7	*ring1a bmi1c*	298.1	2.16	1.52
Treatment	8	*Col-0*	114.7	2.22	2.25
	9	*clf-29*	220.8	2.22	2.33
	10	*swn-3*	347.3	2.19	2.32
	11	*gun1*	447.4	2.21	2.09
	12	*pkl pkr2*	216.3	2.25	2.54
	13	*ring1a bmi1a*	152.3	2.29	2.60
	14	*ring1a bmi1c*	281.6	2.23	2.28

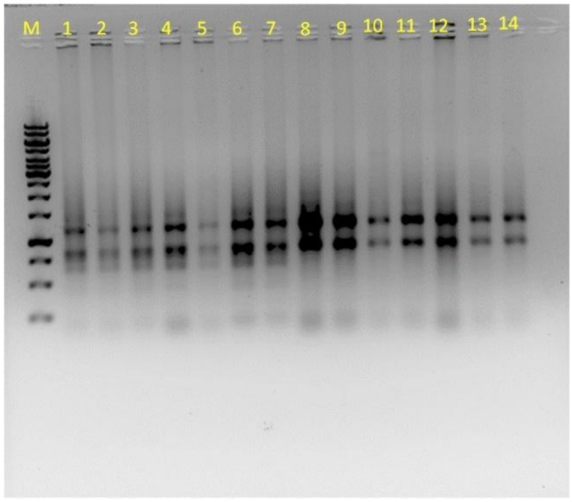

Figure 5: Left panel shows a summary of all samples - amount of isolated total RNA and indication of quality based on the A260/280 and A260/230 ratios. Right image shows results of agarose gel electrophoresis showing RNA bands with different intensities and smearing. The wells on the agarose gel were labelled from 'Marker (M) 1 kb' to 'Well number 14,' corresponding to the plant ID and name. Wells numbered 1 to 7 represent control plant lines that did not receive lincomycin treatment, whereas wells numbered 8 to 14 represent plant lines that cultivated in the presence of lincomycin.

4.3 Analysis of gene transcription using real-time PCR

Reverse transcription quantitative PCR (RT-qPCR) analysis was performed to assess the relative gene transcription levels of the photosynthesis-associated nuclear genes in sets 1 to 4 of control and mutant plants.

4.3.1 Relative gene expression in set 1 of mutant plants

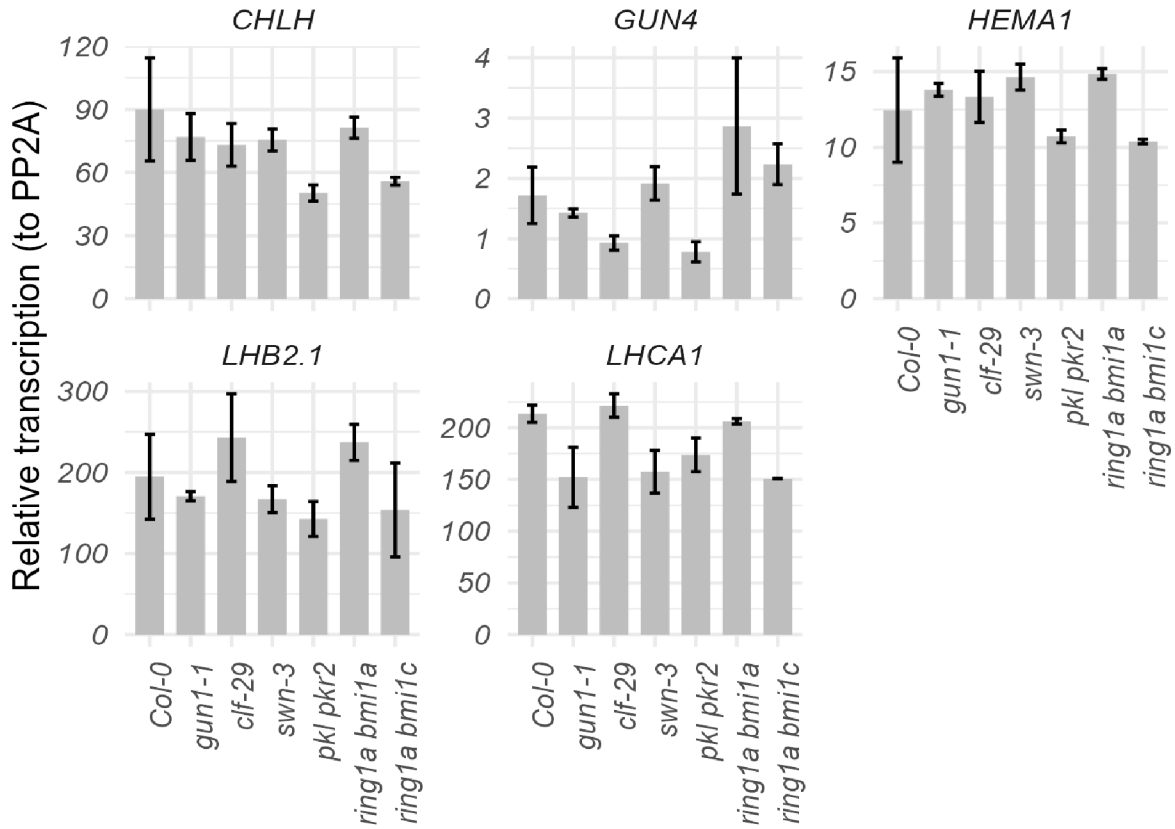

Figure 6: relative gene expression levels of PhANGs under control conditions. Bars indicate relative transcription level, related to the transcription of the marker gene PP2A. Error bars indicate standard deviation of three technical replicates.

Under control conditions, the expression of several PhANGs appeared to be changed in several of the mutant lines. Based on their expression pattern, they seemed to be separated into three groups: 1. *CHLH* and *HEMA* downregulated in *pkl pkr2* and *ring1a bmi1c* compared to Col-0 (WT)*;* 2. *LHCA1* and *LHCB2.1* that tend to be downregulated in *gun1-1*, *swn-3*, *pkl pkr2* and *ring1a bmi1c* and 3. *GUN4* downregulated in *clf-29* and *pkl pkr2.*

Next, we asked whether the gene transcription is reduced upon chloroplast damage by lincomycin, as would be expected in wild-type plants (Figure 7). In line with expectation, the expression of the *PhANG*s decreased in wild type (Col-0) to 5% (*GUN4*) – 20% (*CHLH*) of the transcription level in mock-treated plants. In contrast, the reduction of transcription level reached only ca 25% (*LHCB2.1*) to 60% (*GUN4*) of mock-treated level in *gun1-1*. This indicated that the experimental system works as expected – i.e. *gun1-1* mutants are characterised by less efficient downregulation of *PhANGs* upon lincomycin treatment. Having established the experimental system, we next asked about the presence of *gun* phenotype in the chromatin mutants. In line with the lab´s preliminary results, PRC2 *clf-29* and

especially *swn-3* displayed gun phenotype at least for some genes – namely *CHLH* and *GUN4*. A weak phenotype was observed in the PRC1 mutants *ring1a bmi1a* and *ring1a bmi1c* for the *CHLH* gene. Since the strongest effect was observed when testing the transcription of *CHLH* and *GUN4*, these genes were next selected as marker genes for gun phenotype for the other tested chromatin mutants.

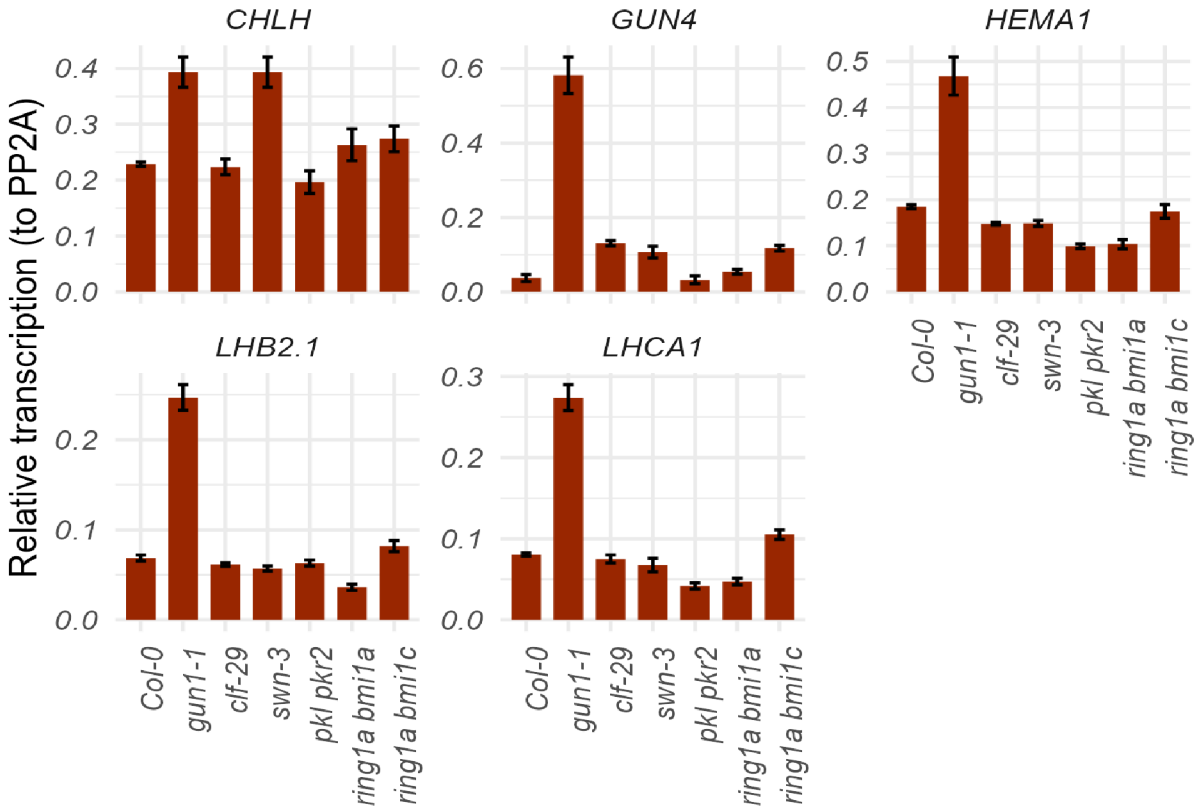

Figure 7: relative gene expression levels of selected PhANGs in plants of set 1 under lincomycin treatment. Bars represent relative transcription values related to control plants (set as 1). Error bars indicate standard deviation of three technical replicates.

4.3.2. Relative gene expression in sets 2, 3 and 4 of mutant plants

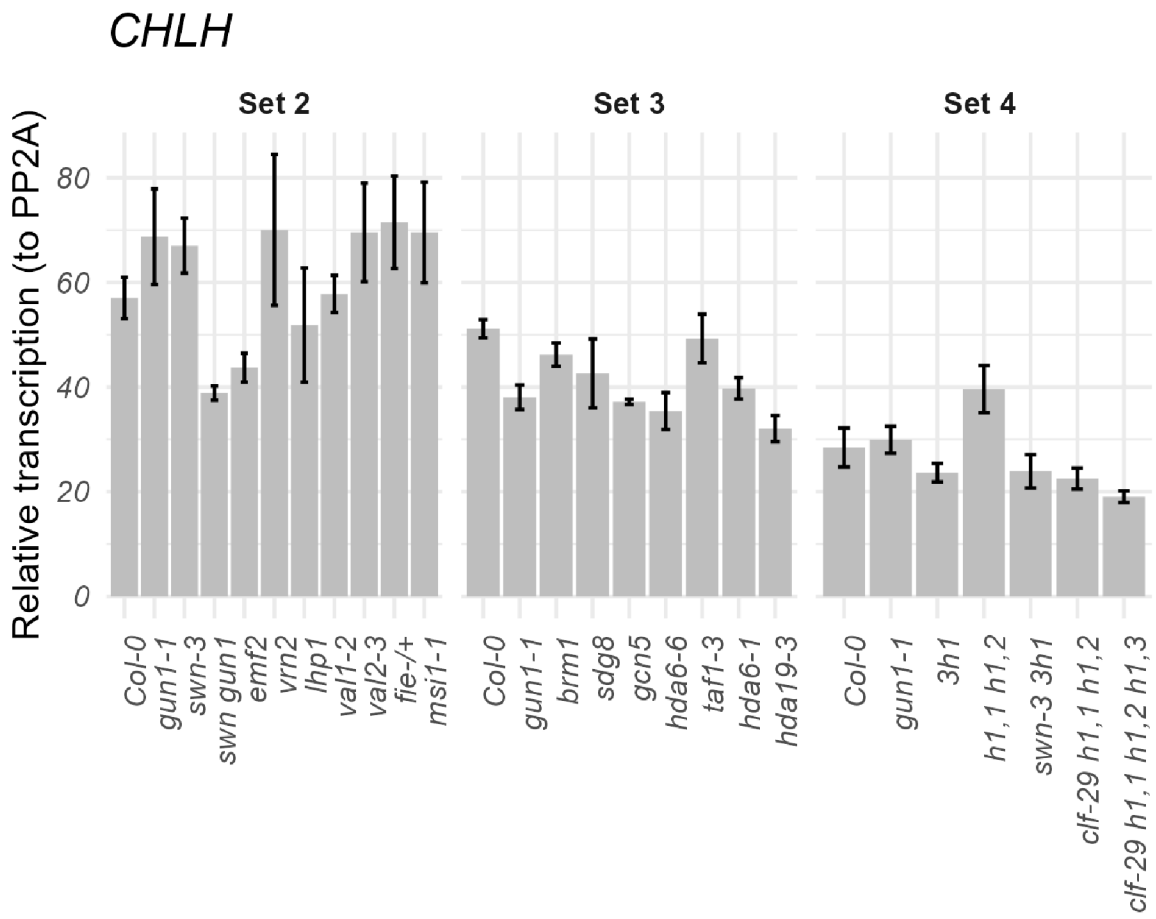

Figure 8: relative gene expression levels of CHLH under control conditions. Bars indicate relative transcription level, related to the transcription of the marker gene PP2A. Error bars indicate standard deviation of three technical replicates.

Under control conditions, the expression of *CHLH* appeared to vary between mutant lines (Figure 8). *CHLH* expression pattern revealed a downregulation in the *swn-3, emf2, gun1-1,* and *3h1* lines, while an upregulation was observed in the *h1,1h1,2* line when compared to the Col-0 control.

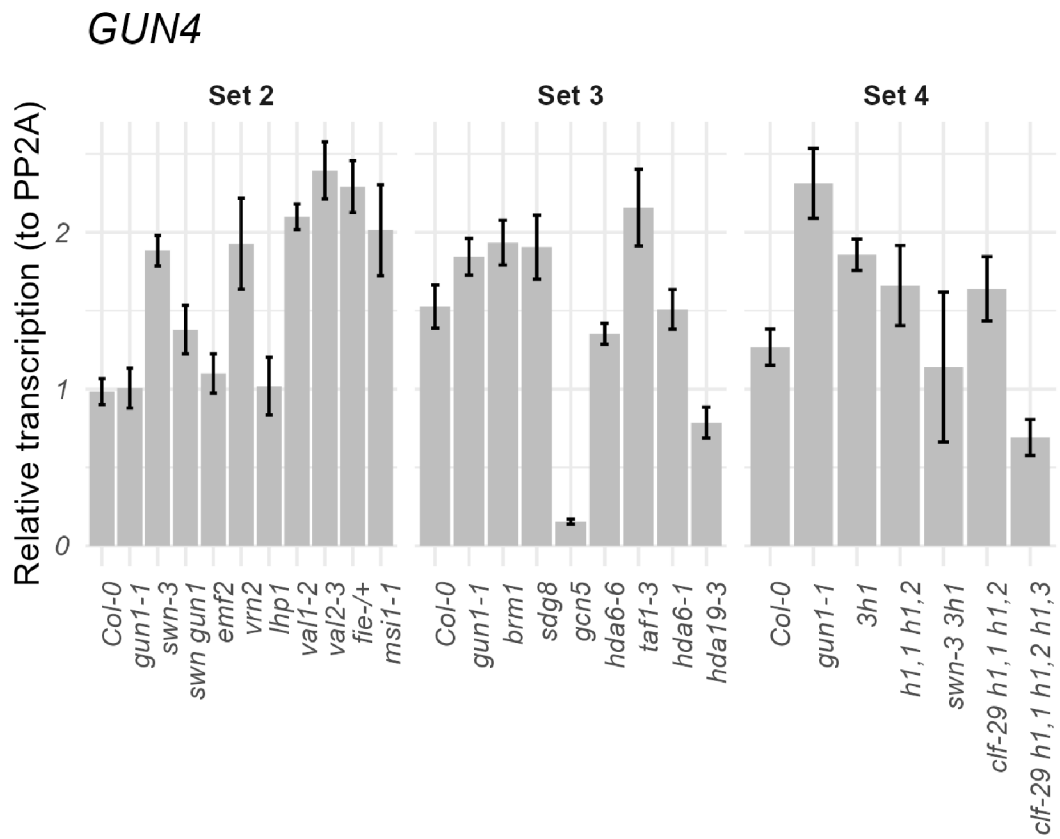

Figure 9: relative gene expression levels of GUN4 under control conditions. Bars indicate relative transcription level, related to the transcription of the marker gene PP2A. Error bars indicate standard deviation of three technical replicates.

Similarly, for GUN4, changes in expression levels under control conditions were apparent in several mutant lines. Based on the observed expression patterns, the *swn-3, vrn2, val1-2, val1-3, fie -/+, msi1-1, taf1-3*, and *gun1-1* lines appeared to be upregulated in comparison to the Col-0. On the contrary, the gcn5, *hda19-3*, and *clf-29 h1,1 h1,2 h1,3* lines exhibited downregulation (Figure 9).

While wild-type (Col-0) plants responded to lincomycin treatment by downregulating the expression of *GUN4*, *gun1-1* mutant plants failed to reach similar level of downregulation (Figure 10). Therefore, control experiments in sets 2 and 3 gave expected results, allowing the interpretation of the *GUN4* transcription level in other tested mutants.

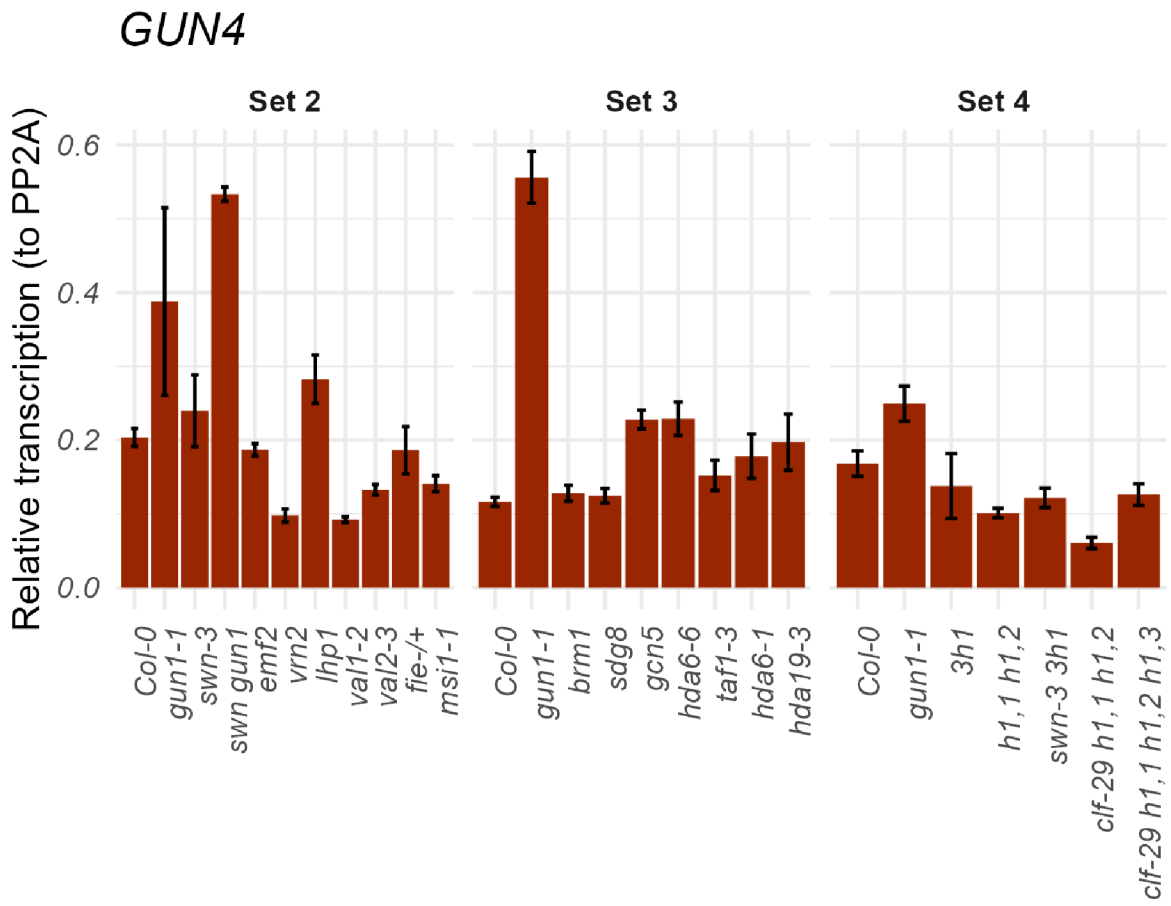

Figure 10: Relative gene expression levels of GUN4 in set 2 to set 4 of chromatin mutants treated with lincomycin. Bars indicate transcription level related to lincomycin-grown plants; error bars indicate standard deviation of three technical replicates. Col-0 (WT) and gun1 served as negative and positive controls, respectively.

While wild-type (Col-0) plants responded to lincomycin treatment by downregulating the expression of *GUN4*, *gun1-1* mutant plants failed to reach similar level of downregulation. Therefore, control experiments in sets 2 and 3 gave expected results, allowing the interpretation of the *GUN4* transcription level in other tested mutants. One exception was set 4, where the difference of *GUN4* transcription after lincomycin treatment between Col-0 and *gun1-1* was in the expected direction (i.e. lower in Col-0), but it was relatively low compared to other sets, although in expected direction. Thus, care must be taken when interpreting the results of set 4. Several mutants in sets 2 and 3 exhibited upregulation of *GUN4* expression in response to lincomycin treatment (Figure 10). In addition to already mentioned mutants in set 1, including both PRC2 mutants (*clf-29* and *swn-3*), and the PRC1 double mutant (*ring1a bmi1c*), the double mutant *swn-3 gun1* and *lhp1* from set 2, and *gcn5*, *hda6-6* and *hda19-3* from set 3, showed

increased transcription levels of *GUN4* compared to the wild-type control. Notably, none of the mutants in set 4 showed *GUN4* upregulation.

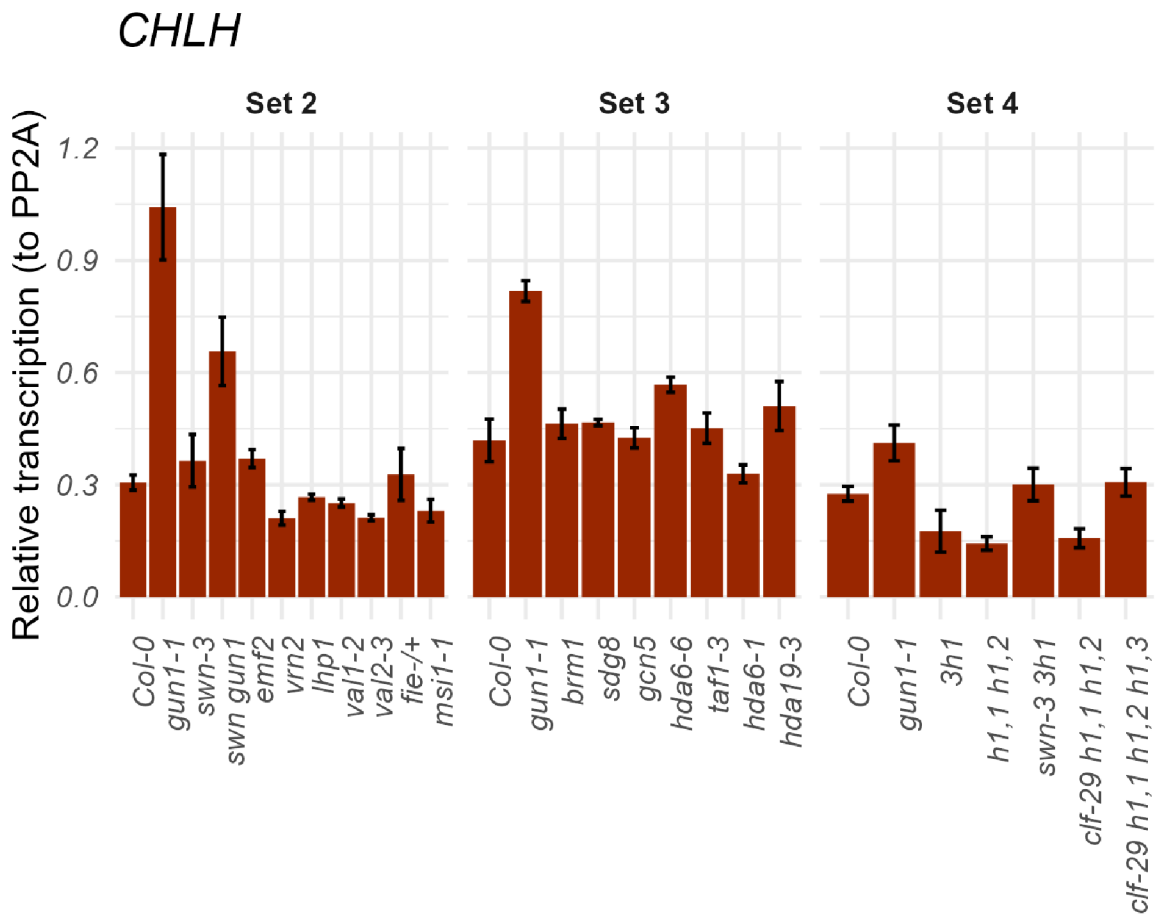

Figure 11: Relative gene expression levels of CHLH in lincomycin treated seedlings of set 2 to set 4 of chromatin mutant plants. Bars indicate transcription level related to lincomycin-grown plants; error bars indicate standard deviation of three technical replicates. Col-0 (WT) and gun1 served as negative and positive controls, respectively.

As for *GUN4*, the transcription of *CHLH* in Col-0 and *gun1-1* followed the expected trend but milder extent of difference was observed in plants of set 4. Both PRC2 mutants (*clf-29* and *swn-3*), and the PRC1 double mutant (*ring1a bmi1c*) demonstrated upregulation of *CHLH* transcription compared to the wild-type control under lincomycin treatment (set 1, Figure 7). Only the double mutant *swn gun1* and *emf2* mutants (set 2) showed a substantial increase in *CHLH* transcription relative to the wild type under lincomycin treatment. In set 3, only the *hda6-6* mutant displayed significant upregulation of *CHLH* transcription compared to the wild-

type under lincomycin treatment. None of the mutants in Set 4 showed upregulated *CHLH* transcription compared to the wild-type control under lincomycin treatment.

Overall, the relative gene expression analysis of *GUN4* and *CHLH* in different mutant sets under lincomycin treatment revealed distinct patterns of upregulation. In set 1, PRC2 mutants (*clf-29* and *swn-3*), and the PRC1 double mutant (*ring1a bmi1c*) displayed significant upregulation of both *GUN4* and *CHLH* compared with the wild type (control). Set 2 showed the upregulation of both genes in *swn gun1 (*double mutant), whereas only *CHLH* showed increased transcription in the *emf2* mutant. In set 3, the HISTONE DEACETYLASE family mutant *hda6-6* exhibited upregulated expression of both *GUN4* and *CHLH*. However, none of the mutants in set 4 showed upregulated gene transcription of either *GUN4* or *CHLH* compared with the wild-type control under lincomycin treatment.

6. Discussion

5.1 Phenotype of seedlings treated with or without lincomycin.

Lincomycin treatment inhibits the translation of plastid genes and alters chloroplast development, resulting in a distinct phenotype with reduced chlorophyll accumulation and cotyledon expansion (Nott et al., 2006). Figure 3 shows that under control conditions, all the seedlings in set 1 had similar growth, chlorophyll levels, and cotyledon size. However, in mature Arabidopsis, *swn-3* had a higher chlorophyll content than WT and *clf-29* (Mingxi Zhou, personal communication). In contrast, lincomycin-treated plant lines exhibited, as expected, deficiency in chlorophyll accumulation and a decrease in cotyledon expansion compared to the control plants. This phenotype was consistent in all seedlings, indicating comparable impairment in chloroplast biogenesis. Under lincomycin treatment, the wild-type had a more pinkish color than other mutants under the same conditions, which might be associated with different anthocyanin biosynthesis in general or under lincomycin treatment. Inhibition of chloroplast biogenesis by lincomycin treatment led to the pale phenotype (Figure 3). This phenotypic change alters retrograde signalling that modulates the expression of nuclear genes involved in various stress responses and developmental processes. Moreover, this phenotype (the pink and pale colour) provided a quick way to extrapolate the efficacy of the added lincomycin without chlorophyll fluorescence analyses. Notably, despite the inhibition of greening in lincomycin-treated plants, the plants were alive and good quality intact RNA was isolated (Figure 3).

5.2 The role of histone modifications in photosynthesis efficiency

Histone modifications, such as methylation and acetylation, are key epigenetic mechanisms that plants use to regulate gene expression in response to developmental signals and environmental stress (Pikaard & Mittelsten Scheid, 2014). Our results showed that the PRC2 mutants (*clf-29* and *swn-3*), as well as the double mutant of PRC1 (*ring1a bmi1c*) and the mutant of the HISTONE DEACETYLASE family *hda6-6*, exhibited upregulation of *GUN4* and *CHLH* genes under lincomycin treatment. *GUN4* is known to be involved in chlorophyll biosynthesis and is a component of retrograde signaling, where it potentially regulates gene expression in response to chloroplast perturbations (Larkin et al., 2003). *CHLH* (*GUN5*) is also involved in retrograde signaling, chlorophyll synthesis, and ABA responses. It is the largest subunit of Mg-chelatase in plastids, catalyzing the conversion of Proto to MgProto, which is a

key step in plant tetrapyrrole biosynthesis (Ibata et al., 2016; Tanaka et al., 2011). The upregulation of these genes suggests an active retrograde signaling pathway in response to lincomycin-induced stress, which may affect chloroplast function.

The Fv/Fm ratio is a sensitive indicator of the maximum quantum efficiency of PSII (Baker, 2008). The uniform Fv/Fm ratios across all plant lines under control conditions suggest that the baseline efficiency of PSII was not significantly affected by the changes in histone modifications under non-stress conditions.

The role of PRC2 in the suppression of gene expression through H3K27me3 is well established for developmentally-regulated genes, and mutations in this complex can also lead to de-repression of genes that may be critical for stress responses (Kleinmanns & Schubert, 2014; Marasca et al., 2018). Following lincomycin treatment, the upregulation of *GUN4* and *CHLH* in PRC2 mutants (*clf-29* and *swn-3*) suggests that PRC2 may repress these genes under normal conditions to enhance photosynthesis efficiency (Figure 6; Figure 7). However, under control conditions, these genes remain largely unchanged in PRC2 mutants. This finding implies that PRC2 may not directly repress *GUN4* and *CHLH*, as their transcription levels are not upregulated in the absence of PRC2, contrary to what would be expected if these genes were under direct repression. The upregulation of these genes in response to lincomycin treatment suggests a potentially more complex role for PRC2, possibly modulating stress responses associated with chloroplast dysfunction, rather than acting as a simple repressor. Furthermore, the upregulation of *clf-29* and *swn-3* mutants in *GUN4* and *CHLH* under lincomycin conditions suggests that PRC2 may play a role in retrograde signaling pathways. This implies that the histone methylation status, regulated by PRC2, could be a significant factor in controlling the expression of genes involved in retrograde signaling. The role of PRC1 in plant development and its potential involvement in stress responses have been reported in the literature (Bratzel et al., 2010). The upregulation of photosynthesis-related genes in the double mutant *ring1a bmi1c* could indicate a de-repressive mechanism analogous to that observed in PRC2 mutants, contributing to the maintenance of PSII efficiency under stress conditions.

The *hda6-6* mutant also exhibits upregulation of both *GUN4* and *CHLH* after lincomycin treatment. This is consistent with the known function of HDA6 in repressing gene expression by histone deacetylation (L.-T. Chen et al., 2010). Loss of function in *hda6-6* could lead to hyperacetylation and active transcription of these genes, potentially enhancing the plant's ability to respond to stress induced by lincomycin by promoting PSII function. The ability to

modulate photosynthesis-related gene expression through epigenetic mechanisms under stress conditions can be considered an evolutionary advantage, allowing plants to survive and reproduce in fluctuating environments (Chinnusamy & Zhu, 2009).

Epigenetic modifications allow plants to dynamically adjust gene expression according to internal cues that indicate the physiological status of chloroplasts. These alterations in gene expression patterns facilitate the adaptation of plants to stressful conditions, preserve chloroplast function, and ensure cellular balance. These findings highlight the intricacy of epigenetic regulation of retrograde signaling. Histone modifications play a crucial role in influencing this communication between chloroplasts and the nucleus, especially under stress conditions that impair chloroplast function and require adaptive responses, such as retrograde signaling pathways, to sustain plant viability.

7. Conclusions

The findings of this thesis highlight the significant role of histone modifications in mediating the response of *Arabidopsis thaliana* to light acclimation through retrograde signalling. Specific histone modification mutants, including those in the PRC2 and PRC1 complexes, as well as the HISTONE DEACETYLASE family, have been shown to exhibit differential regulation of genes essential to these processes, such as *GUN4* and *CHLH*. The upregulation of these genes in response to lincomycin treatment indicates an adaptive mechanism facilitated by epigenetic modulation, which is critical for maintaining photosynthetic efficiency under stress conditions.

The role of histone modifications in photosynthetic efficiency is underscored by the maintenance of PSII functionality, as reflected in the Fv/Fm ratio data. Despite the uniform Fv/Fm ratios observed under control conditions, the stress response induced by lincomycin treatment was characterized by a reduction in PSII efficiency across all plant lines. The upregulation of these genes in histone modification mutants underlines the importance of chromatin remodelling in response to retrograde signalling. Histone modifications have been implicated in modulating the expression of genes that facilitate communication between the chloroplast and nucleus, thereby influencing the plant's ability to acclimate to changes in light conditions. This dynamic regulation is pivotal for plant adaptation, ensuring that photosynthesis remains efficient, and that cellular homeostasis is preserved during environmental stress. The results presented were based on the relative gene expression levels of one biological replicate with three technical replicates, hence no statistical analyses was conducted. However, the data provide valuable preliminary information on the role of chromatin modifiers in plant acclimation to light, setting the foundation for future studies that should aim to include a larger number of biological replicates to validate the findings reported in this thesis.

These observations contribute to a broader understanding of the molecular mechanisms underlying plant light acclimation and highlight the potential of epigenetic modifications as targets for genetic or chemical modulation to improve crop resilience and productivity in the face of changing environmental conditions.

8. Literature

Adhikari, N. D., Froehlich, J. E., Strand, D. D., Buck, S. M., Kramer, D. M., & Larkin, R. M. (2011). GUN4-Porphyrin Complexes Bind the ChlH/GUN5 Subunit of Mg-Chelatase and Promote Chlorophyll Biosynthesis in Arabidopsis. The Plant Cell, 23(4), 1449–1467. https://doi.org/10.1105/tpc.110.082503

Alberts, B., Johnson, A., Lewis, J., Raff, M., Roberts, K., & Walter, P. (2002). Molecular Biology of the Cell (4th ed.). Garland Science.

Archibald, J. M. (2015). Endosymbiosis and Eukaryotic Cell Evolution. Current Biology, 25(19), R911–R921. https://doi.org/10.1016/j.cub.2015.07.055

Arsovski, A. A., Galstyan, A., Guseman, J. M., & Nemhauser, J. L. (2012). Photomorphogenesis. The Arabidopsis Book / American Society of Plant Biologists, 10, e0147. https://doi.org/10.1199/tab.0147

Bae, G., & Choi, G. (2008). Decoding of Light Signals by Plant Phytochromes and Their Interacting Proteins. Annual Review of Plant Biology, 59(1), 281–311. https://doi.org/10.1146/annurev.arplant.59.032607.092859

Baile, F., Gómez-Zambrano, Á., & Calonje, M. (2022). Roles of Polycomb complexes in regulating gene expression and chromatin structure in plants. Plant Communications, 3(1). https://doi.org/10.1016/j.xplc.2021.100267

Baker, N. R. (2008). Chlorophyll Fluorescence: A Probe of Photosynthesis In Vivo. Annual Review of Plant Biology, 59(1), 89–113. https://doi.org/10.1146/annurev.arplant.59.032607.092759

Bao, Y., & Howell, S. H. (2017). The Unfolded Protein Response Supports Plant Development and Defense as well as Responses to Abiotic Stress. Frontiers in Plant Science, 8. https://www.frontiersin.org/articles/10.3389/fpls.2017.00344

Barajas-López, J. de D., Blanco, N. E., & Strand, Å. (2013). Plastid-to-nucleus communication, signals controlling the running of the plant cell. Biochimica et Biophysica Acta (BBA) - Molecular Cell Research, 1833(2), 425–437. https://doi.org/10.1016/j.bbamcr.2012.06.020

Bastakis, E., Hedtke, B., Klermund, C., Grimm, B., & Schwechheimer, C. (2018). LLM-Domain B-GATA Transcription Factors Play Multifaceted Roles in Controlling Greening in Arabidopsis. The Plant Cell, 30(3), 582–599. https://doi.org/10.1105/tpc.17.00947

Bastien, O., Botella, C., Chevalier, F., Block, M. A., Jouhet, J., Breton, C., Girard-Egrot, A., & Maréchal, E. (2016). Chapter One—New Insights on Thylakoid Biogenesis in Plant Cells. In K. W.

Jeon (Ed.), International Review of Cell and Molecular Biology (Vol. 323, pp. 1–30). Academic Press. https://doi.org/10.1016/bs.ircmb.2015.12.001

Beisel, C., & Paro, R. (2011). Silencing chromatin: Comparing modes and mechanisms. Nature Reviews Genetics, 12(2), Article 2. https://doi.org/10.1038/nrg2932

Benhamed, M., Bertrand, C., Servet, C., & Zhou, D.-X. (2006). Arabidopsis GCN5, HD1, and TAF1/HAF2 Interact to Regulate Histone Acetylation Required for Light-Responsive Gene Expression. The Plant Cell, 18(11), 2893–2903. https://doi.org/10.1105/tpc.106.043489

Berger, S. L., Kouzarides, T., Shiekhattar, R., & Shilatifard, A. (2009). An operational definition of epigenetics. Genes & Development, 23(7), 781–783. https://doi.org/10.1101/gad.1787609

Berke, L., & Snel, B. (2015). The plant Polycomb repressive complex 1 (PRC1) existed in the ancestor of seed plants and has a complex duplication history. BMC Evolutionary Biology, 15, 44. https://doi.org/10.1186/s12862-015-0319-z

Bertrand, C., Bergounioux, C., Domenichini, S., Delarue, M., & Zhou, D.-X. (2003). Arabidopsis Histone Acetyltransferase AtGCN5 Regulates the Floral Meristem Activity through the WUSCHEL/AGAMOUS Pathway*. Journal of Biological Chemistry, 278(30), 28246–28251. https://doi.org/10.1074/jbc.M302787200

Bian, Z., Gao, H., & Wang, C. (2020). NAC Transcription Factors as Positive or Negative Regulators during Ongoing Battle between Pathogens and Our Food Crops. International Journal of Molecular Sciences, 22(1), 81. https://doi.org/10.3390/ijms22010081

Börner, T., Aleynikova, A. Y., Zubo, Y. O., & Kusnetsov, V. V. (2015). Chloroplast RNA polymerases: Role in chloroplast biogenesis. Biochimica Et Biophysica Acta, 1847(9), 761–769. https://doi.org/10.1016/j.bbabio.2015.02.004

Bratzel, F., López-Torrejón, G., Koch, M., Pozo, J. C. D., & Calonje, M. (2010). Keeping Cell Identity in Arabidopsis Requires PRC1 RING-Finger Homologs that Catalyze H2A Monoubiquitination. Current Biology, 20(20), 1853–1859. https://doi.org/10.1016/j.cub.2010.09.046

Cackett, L., Luginbuehl, L. H., Schreier, T. B., Lopez-Juez, E., & Hibberd, J. M. (2022). Chloroplast development in green plant tissues: The interplay between light, hormone, and transcriptional regulation. New Phytologist, 233(5), 2000–2016. https://doi.org/10.1111/nph.17839

Calonje, M. (2014). PRC1 Marks the Difference in Plant PcG Repression. Molecular Plant, 7(3), 459–471. https://doi.org/10.1093/mp/sst150

Chaffey, N. (2014). Raven biology of plants, 8th edn. Annals of Botany, 113(7), vii. https://doi.org/10.1093/aob/mcu090

Chan, K. X., Phua, S. Y., Crisp, P., McQuinn, R., & Pogson, B. J. (2016). Learning the Languages of the Chloroplast: Retrograde Signaling and Beyond. Annual Review of Plant Biology, 67(1), 25–53. https://doi.org/10.1146/annurev-arplant-043015-111854

Chen, L.-T., Luo, M., Wang, Y.-Y., & Wu, K. (2010). Involvement of Arabidopsis histone deacetylase HDA6 in ABA and salt stress response. Journal of Experimental Botany, 61(12), 3345–3353. https://doi.org/10.1093/jxb/erq154

Chen, L.-T., & Wu, K. (2010). Role of histone deacetylases HDA6 and HDA19 in ABA and abiotic stress response. Plant Signaling & Behavior, 5(10), 1318–1320. https://doi.org/10.4161/psb.5.10.13168

Chen, M., Galvão, R. M., Li, M., Burger, B., Bugea, J., Bolado, J., & Chory, J. (2010). Arabidopsis HEMERA/pTAC12 Initiates Photomorphogenesis by Phytochromes. Cell, 141(7), 1230–1240. https://doi.org/10.1016/j.cell.2010.05.007

Chi, W., He, B., Mao, J., Jiang, J., & Zhang, L. (2015). Plastid sigma factors: Their individual functions and regulation in transcription. Biochimica et Biophysica Acta (BBA) - Bioenergetics, 1847(9), 770–778. https://doi.org/10.1016/j.bbabio.2015.01.001

Chi, W., Mao, J., Li, Q., Ji, D., Zou, M., Lu, C., & Zhang, L. (2010). Interaction of the pentatricopeptide-repeat protein DELAYED GREENING 1 with sigma factor SIG6 in the regulation of chloroplast gene expression in Arabidopsis cotyledons. The Plant Journal, 64(1), 14–25. https://doi.org/10.1111/j.1365-313X.2010.04304.x

Chi, W., Sun, X., & Zhang, L. (2013). Intracellular Signaling from Plastid to Nucleus. Annual Review of Plant Biology, 64(1), 559–582. https://doi.org/10.1146/annurev-arplant-050312-120147

Chinnusamy, V., & Zhu, J.-K. (2009). Epigenetic regulation of stress responses in plants. Current Opinion in Plant Biology, 12(2), 133–139. https://doi.org/10.1016/j.pbi.2008.12.006

Chory, J., Peto, C. A., Ashbaugh, M., Saganich, R., Pratt, L., & Ausubel, F. (1989). Different Roles for Phytochrome in Etiolated and Green Plants Deduced from Characterization of Arabidopsis thaliana Mutants. The Plant Cell, 1(9), 867–880. https://doi.org/10.2307/3868934

Christian, R. W., Hewitt, S. L., Roalson, E. H., & Dhingra, A. (2020). Genome-Scale Characterization of Predicted Plastid-Targeted Proteomes in Higher Plants. Scientific Reports, 10(1), Article 1. https://doi.org/10.1038/s41598-020-64670-5

Clapier, C. R., & Cairns, B. R. (2009). The Biology of Chromatin Remodeling Complexes. Annual Review of Biochemistry, 78(1), 273–304. https://doi.org/10.1146/annurev.biochem.77.062706.153223

Colombo, M., Tadini, L., Peracchio, C., Ferrari, R., & Pesaresi, P. (2016). GUN1, a Jack-Of-All-Trades in Chloroplast Protein Homeostasis and Signaling. Frontiers in Plant Science, 7. https://doi.org/10.3389/fpls.2016.01427

Crawford, T., Lehotai, N., & Strand, Å. (2018). The role of retrograde signals during plant stress responses. Journal of Experimental Botany, 69(11), 2783–2795. https://doi.org/10.1093/jxb/erx481

Crevillén, P. (2020). Histone Demethylases as Counterbalance to H3K27me3 Silencing in Plants. iScience, 23(11), 101715. https://doi.org/10.1016/j.isci.2020.101715

Crevillén, P., Yang, H., Cui, X., Greeff, C., Trick, M., Qiu, Q., Cao, X., & Dean, C. (2014). Epigenetic reprogramming that prevents transgenerational inheritance of the vernalized state. Nature, 515(7528), 587–590. https://doi.org/10.1038/nature13722

Daniell, H., Lin, C.-S., Yu, M., & Chang, W.-J. (2016). Chloroplast genomes: Diversity, evolution, and applications in genetic engineering. Genome Biology, 17(1), 134. https://doi.org/10.1186/s13059-016-1004-2

Day, P. M., & Theg, S. M. (2018). Evolution of protein transport to the chloroplast envelope membranes. Photosynthesis Research, 138(3), 315–326. https://doi.org/10.1007/s11120-018-0540-x

De Lucia, F., Crevillen, P., Jones, A. M. E., Greb, T., & Dean, C. (2008). A PHD-Polycomb Repressive Complex 2 triggers the epigenetic silencing of FLC during vernalization. Proceedings of the National Academy of Sciences of the United States of America, 105(44), 16831–16836. https://doi.org/10.1073/pnas.0808687105

de Souza, A., Wang, J.-Z., & Dehesh, K. (2017). Retrograde Signals: Integrators of Interorganellar Communication and Orchestrators of Plant Development. Annual Review of Plant Biology, 68(1), 85–108. https://doi.org/10.1146/annurev-arplant-042916-041007

Deng, W., Liu, C., Pei, Y., Deng, X., Niu, L., & Cao, X. (2007). Involvement of the Histone Acetyltransferase AtHAC1 in the Regulation of Flowering Time via Repression of FLOWERING LOCUS C in Arabidopsis. Plant Physiology, 143(4), 1660–1668. https://doi.org/10.1104/pp.107.095521

Díaz, M. G., Hernández-Verdeja, T., Kremnev, D., Crawford, T., Dubreuil, C., & Strand, Å. (2018). Redox regulation of PEP activity during seedling establishment in Arabidopsis thaliana. Nature Communications, 9(1), 50. https://doi.org/10.1038/s41467-017-02468-2

Dobrogojski, J., Adamiec, M., & Luciński, R. (2020). The chloroplast genome: A review. Acta Physiologiae Plantarum, 42(6), 98. https://doi.org/10.1007/s11738-020-03089-x

Espinas, N. A., Saze, H., & Saijo, Y. (2016). Epigenetic Control of Defense Signaling and Priming in Plants. Frontiers in Plant Science, 7. https://www.frontiersin.org/articles/10.3389/fpls.2016.01201

Fang, W., Fasano, C., & Perrella, G. (2023). Unlocking the Secret to Higher Crop Yield: The Potential for Histone Modifications. Plants, 12(8), Article 8. https://doi.org/10.3390/plants12081712

Fitter, D. W., Martin, D. J., Copley, M. J., Scotland, R. W., & Langdale, J. A. (2002). GLK gene pairs regulate chloroplast development in diverse plant species. The Plant Journal, 31(6), 713–727. https://doi.org/10.1046/j.1365-313X.2002.01390.x

Floris, D., & Kühlbrandt, W. (2021). Molecular landscape of etioplast inner membranes in higher plants. Nature Plants, 7(4), Article 4. https://doi.org/10.1038/s41477-021-00896-z

Francis, N. J., Kingston, R. E., & Woodcock, C. L. (2004). Chromatin Compaction by a Polycomb Group Protein Complex. Science, 306(5701), 1574–1577. https://doi.org/10.1126/science.1100576

Fu, X.-X., Zhang, J., Zhang, G.-Q., Liu, Z.-J., & Chen, Z.-D. (2021). Insights into the origin and evolution of plant sigma factors. Journal of Systematics and Evolution, 59(2), 326–340. https://doi.org/10.1111/jse.12537

Fyodorov, D. V., Zhou, B.-R., Skoultchi, A. I., & Bai, Y. (2018). Emerging roles of linker histones in regulating chromatin structure and function. Nature Reviews Molecular Cell Biology, 19(3), Article 3. https://doi.org/10.1038/nrm.2017.94

Gadjieva, R., Axelsson, E., Olsson, U., & Hansson, M. (2005). Analysis of gun phenotype in barley magnesium chelatase and Mg-protoporphyrin IX monomethyl ester cyclase mutants. Plant Physiology and Biochemistry: PPB, 43(10–11), 901–908. https://doi.org/10.1016/j.plaphy.2005.08.003

Galvão, R. M., Li, M., Kothadia, S. M., Haskel, J. D., Decker, P. V., Buskirk, E. K. V., & Chen, M. (2012). Photoactivated phytochromes interact with HEMERA and promote its accumulation to establish photomorphogenesis in Arabidopsis. Genes & Development, 26(16), 1851–1863. https://doi.org/10.1101/gad.193219.112

Gao, Z.-P., Yu, Q.-B., Zhao, T.-T., Ma, Q., Chen, G.-X., & Yang, Z.-N. (2011). A Functional Component of the Transcriptionally Active Chromosome Complex, Arabidopsis pTAC14, Interacts with pTAC12/HEMERA and Regulates Plastid Gene Expression. Plant Physiology, 157(4), 1733–1745. https://doi.org/10.1104/pp.111.184762

Gawroński, P., Burdiak, P., Scharff, L. B., Mielecki, J., Górecka, M., Zaborowska, M., Leister, D., Waszczak, C., & Karpiński, S. (2021). CIA2 and CIA2-LIKE are required for optimal photosynthesis and stress responses in Arabidopsis thaliana. The Plant Journal, 105(3), 619–638. https://doi.org/10.1111/tpj.15058

Gibney, E. R., & Nolan, C. M. (2010). Epigenetics and gene expression. Heredity, 105(1), Article 1. https://doi.org/10.1038/hdy.2010.54

Gläßer, C., Haberer, G., Finkemeier, I., Pfannschmidt, T., Kleine, T., Leister, D., Dietz, K.-J., Häusler, R. E., Grimm, B., & Mayer, K. F. X. (2014). Meta-Analysis of Retrograde Signaling in Arabidopsis thaliana Reveals a Core Module of Genes Embedded in Complex Cellular Signaling Networks. Molecular Plant, 7(7), 1167–1190. https://doi.org/10.1093/mp/ssu042

Gommers, C. M. M., Ruiz-Sola, M. Á., Ayats, A., Pereira, L., Pujol, M., & Monte, E. (2020). GENOMES UNCOUPLED1-independent retrograde signaling targets the ethylene pathway to repress photomorphogenesis. Plant Physiology, 185(1), 67–76. https://doi.org/10.1093/plphys/kiaa015

Goodrich, J., Puangsomlee, P., Martin, M., Long, D., Meyerowitz, E. M., & Coupland, G. (1997). A Polycomb-group gene regulates homeotic gene expression in Arabidopsis. Nature, 386(6620), Article 6620. https://doi.org/10.1038/386044a0

Gray, J. C., Sullivan, J. A., Wang, J.-H., Jerome, C. A., & MacLean, D. (2003). Coordination of plastid and nuclear gene expression. Philosophical Transactions of the Royal Society B: Biological Sciences, 358(1429), 135–145. https://doi.org/10.1098/rstb.2002.1180

Gray, M. W., Burger, G., & Lang, B. F. (1999). Mitochondrial evolution. Science (New York, N.Y.), 283(5407), 1476–1481. https://doi.org/10.1126/science.283.5407.1476

Grübler, B., Merendino, L., Twardziok, S. O., Mininno, M., Allorent, G., Chevalier, F., Liebers, M., Blanvillain, R., Mayer, K. F. X., Lerbs-Mache, S., Ravanel, S., & Pfannschmidt, T. (2017). Light and Plastid Signals Regulate Different Sets of Genes in the Albino Mutant Pap7-1. Plant Physiology, 175(3), 1203–1219. https://doi.org/10.1104/pp.17.00982

Hall, L. N., Rossini, L., Cribb, L., & Langdale, J. A. (1998). GOLDEN 2: A novel transcriptional regulator of cellular differentiation in the maize leaf. The Plant Cell, 10(6), 925–936.

He, K., Mei, H., Zhu, J., Qiu, Q., Cao, X., & Deng, X. (2021). The histone H3K27 demethylase REF6/JMJ12 promotes thermomorphogenesis in Arabidopsis. National Science Review, 9(5), nwab213. https://doi.org/10.1093/nsr/nwab213

Hedtke, B., Börner, T., & Weihe, A. (1997). Mitochondrial and Chloroplast Phage-Type RNA Polymerases in Arabidopsis. Science, 277(5327), 809–811. https://doi.org/10.1126/science.277.5327.809

Hemenway, E. A., & Gehring, M. (2023). Epigenetic Regulation During Plant Development and the Capacity for Epigenetic Memory. Annual Review of Plant Biology, 74(1), 87–109. https://doi.org/10.1146/annurev-arplant-070122-025047

Hernández-Verdeja, T., & Strand, Å. (2018). Retrograde Signals Navigate the Path to Chloroplast Development1[OPEN]. Plant Physiology, 176(2), 967–976. https://doi.org/10.1104/pp.17.01299

Hickman, R., Hill, C., Penfold, C. A., Breeze, E., Bowden, L., Moore, J. D., Zhang, P., Jackson, A., Cooke, E., Bewicke-Copley, F., Mead, A., Beynon, J., Wild, D. L., Denby, K. J., Ott, S., & Buchanan-Wollaston, V. (2013). A local regulatory network around three NAC transcription factors in stress responses and senescence in Arabidopsis leaves. The Plant Journal, 75(1), 26–39. https://doi.org/10.1111/tpj.12194

Hills, A. C., Khan, S., & López-Juez, E. (2015). Chloroplast Biogenesis-Associated Nuclear Genes: Control by Plastid Signals Evolved Prior to Their Regulation as Part of Photomorphogenesis. Frontiers in Plant Science, 6, 1078. https://doi.org/10.3389/fpls.2015.01078

Hricová, A., Quesada, V., & Micol, J. L. (2006). The SCABRA3 Nuclear Gene Encodes the Plastid RpoTp RNA Polymerase, Which Is Required for Chloroplast Biogenesis and Mesophyll Cell Proliferation in Arabidopsis. Plant Physiology, 141(3), 942–956. https://doi.org/10.1104/pp.106.080069

Hudson, D., Guevara, D. R., Hand, A. J., Xu, Z., Hao, L., Chen, X., Zhu, T., Bi, Y.-M., & Rothstein, S. J. (2013). Rice cytokinin GATA transcription Factor1 regulates chloroplast development and plant architecture. Plant Physiology, 162(1), 132–144. https://doi.org/10.1104/pp.113.217265

Ibata, H., Nagatani, A., & Mochizuki, N. (2016). CHLH/GUN5 Function in Tetrapyrrole Metabolism Is Correlated with Plastid Signaling but not ABA Responses in Guard Cells. Frontiers in Plant Science, 7. https://www.frontiersin.org/articles/10.3389/fpls.2016.01650

Inaba, T., Yazu, F., Ito-Inaba, Y., Kakizaki, T., & Nakayama, K. (2011). Chapter Five—Retrograde Signaling Pathway from Plastid to Nucleus. In K. W. Jeon (Ed.), International Review of Cell and

Molecular Biology (Vol. 290, pp. 167–204). Academic Press. https://doi.org/10.1016/B978-0-12-386037-8.00002-8

Jain, K., Fraser, C. S., Marunde, M. R., Parker, M. M., Sagum, C., Burg, J. M., Hall, N., Popova, I. K., Rodriguez, K. L., Vaidya, A., Krajewski, K., Keogh, M.-C., Bedford, M. T., & Strahl, B. D. (2020). Characterization of the plant homeodomain (PHD) reader family for their histone tail interactions. Epigenetics & Chromatin, 13(1), 3. https://doi.org/10.1186/s13072-020-0328-z

Jan, M., Liu, Z., Rochaix, J.-D., & Sun, X. (2022). Retrograde and anterograde signaling in the crosstalk between chloroplast and nucleus. Frontiers in Plant Science, 13. https://www.frontiersin.org/articles/10.3389/fpls.2022.980237

Jarvis, P., & López-Juez, E. (2013). Biogenesis and homeostasis of chloroplasts and other plastids. Nature Reviews Molecular Cell Biology, 14(12), Article 12. https://doi.org/10.1038/nrm3702

Jia, Y., Tian, H., Zhang, S., Ding, Z., & Ma, C. (2019). GUN1-Interacting Proteins Open the Door for Retrograde Signaling. Trends in Plant Science, 24(10), 884–887. https://doi.org/10.1016/j.tplants.2019.07.005

Jiang, J., Zeng, L., Ke, H., De La Cruz, B., & Dehesh, K. (2019). Orthogonal regulation of phytochrome B abundance by stress-specific plastidial retrograde signaling metabolite. Nature Communications, 10(1), Article 1. https://doi.org/10.1038/s41467-019-10867-w

Kacprzak, S. M., Mochizuki, N., Naranjo, B., Xu, D., Leister, D., Kleine, T., Okamoto, H., & Terry, M. J. (2019). Plastid-to-Nucleus Retrograde Signalling during Chloroplast Biogenesis Does Not Require ABI4. Plant Physiology, 179(1), 18–23. https://doi.org/10.1104/pp.18.01047

Kakizaki, T., Matsumura, H., Nakayama, K., Che, F.-S., Terauchi, R., & Inaba, T. (2009). Coordination of Plastid Protein Import and Nuclear Gene Expression by Plastid-to-Nucleus Retrograde Signaling. Plant Physiology, 151(3), 1339–1353. https://doi.org/10.1104/pp.109.145987

Kindgren, P., Dubreuil, C., & Strand, Å. (2015). The Recovery of Plastid Function Is Required for Optimal Response to Low Temperatures in Arabidopsis. PLOS ONE, 10(9), e0138010. https://doi.org/10.1371/journal.pone.0138010

Kindgren, P., Eriksson, M.-J., Benedict, C., Mohapatra, A., Gough, S. P., Hansson, M., Kieselbach, T., & Strand, A. (2011). A novel proteomic approach reveals a role for Mg-protoporphyrin IX in response to oxidative stress. Physiologia Plantarum, 141(4), 310–320. https://doi.org/10.1111/j.1399-3054.2010.01440.x

Kleinmanns, J. A., & Schubert, D. (2014). Polycomb and Trithorax group protein-mediated control of stress responses in plants. Biological Chemistry, 395(11), 1291–1300. https://doi.org/10.1515/hsz-2014-0197

Kobayashi, K., & Masuda, T. (2016). Transcriptional Regulation of Tetrapyrrole Biosynthesis in Arabidopsis thaliana. Frontiers in Plant Science, 7, 1811. https://doi.org/10.3389/fpls.2016.01811

Kohchi, T., Mukougawa, K., Frankenberg, N., Masuda, M., Yokota, A., & Lagarias, J. C. (2001). The Arabidopsis HY2 Gene Encodes Phytochromobilin Synthase, a Ferredoxin-Dependent Biliverdin Reductase. The Plant Cell, 13(2), 425–436. https://doi.org/10.1105/tpc.13.2.425

Köhler, C., Hennig, L., Bouveret, R., Gheyselinck, J., Grossniklaus, U., & Gruissem, W. (2003). Arabidopsis MSI1 is a component of the MEA/FIE Polycomb group complex and required for seed development. The EMBO Journal, 22(18), 4804–4814. https://doi.org/10.1093/emboj/cdg444

Koornneef, M., & Meinke, D. (2010). The development of Arabidopsis as a model plant. The Plant Journal, 61(6), 909–921. https://doi.org/10.1111/j.1365-313X.2009.04086.x

Kornet, N., & Scheres, B. (2009). Members of the GCN5 Histone Acetyltransferase Complex Regulate PLETHORA-Mediated Root Stem Cell Niche Maintenance and Transit Amplifying Cell Proliferation in Arabidopsis. The Plant Cell, 21(4), 1070–1079. https://doi.org/10.1105/tpc.108.065300

Koussevitzky, S., Nott, A., Mockler, T. C., Hong, F., Sachetto-Martins, G., Surpin, M., Lim, J., Mittler, R., & Chory, J. (2007). Signals from Chloroplasts Converge to Regulate Nuclear Gene Expression. Science, 316(5825), 715–719. https://doi.org/10.1126/science.1140516

Krogan, N. T., Hogan, K., & Long, J. A. (2012). APETALA2 negatively regulates multiple floral organ identity genes in Arabidopsis by recruiting the co-repressor TOPLESS and the histone deacetylase HDA19. Development (Cambridge, England), 139(22), 4180–4190. https://doi.org/10.1242/dev.085407

Langdale, J. A., & Kidner, C. A. (1994). Bundle sheath defective, a mutation that disrupts cellular differentiation in maize leaves. Development, 120(3), 673–681. https://doi.org/10.1242/dev.120.3.673

Larkin, R. M. (2014). Influence of plastids on light signalling and development. Philosophical Transactions of the Royal Society B: Biological Sciences, 369(1640), 20130232. https://doi.org/10.1098/rstb.2013.0232

Larkin, R. M., Alonso, J. M., Ecker, J. R., & Chory, J. (2003). GUN4, a regulator of chlorophyll synthesis and intracellular signaling. Science (New York, N.Y.), 299(5608), 902–906. https://doi.org/10.1126/science.1079978

Lee, J., He, K., Stolc, V., Lee, H., Figueroa, P., Gao, Y., Tongprasit, W., Zhao, H., Lee, I., & Deng, X. W. (2007). Analysis of Transcription Factor HY5 Genomic Binding Sites Revealed Its Hierarchical Role in Light Regulation of Development. The Plant Cell, 19(3), 731–749. https://doi.org/10.1105/tpc.106.047688

Leister, D., & Kleine, T. (2016). Definition of a core module for the nuclear retrograde response to altered organellar gene expression identifies GLK overexpressors as gun mutants. Physiologia Plantarum, 157(3), 297–309. https://doi.org/10.1111/ppl.12431

Li, J., Li, G., Wang, H., & Wang Deng, X. (2011). Phytochrome Signaling Mechanisms. The Arabidopsis Book / American Society of Plant Biologists, 9, e0148. https://doi.org/10.1199/tab.0148

Li, M., Hensel, G., Mascher, M., Melzer, M., Budhagatapalli, N., Rutten, T., Himmelbach, A., Beier, S., Korzun, V., Kumlehn, J., Börner, T., & Stein, N. (2019). Leaf Variegation and Impaired Chloroplast Development Caused by a Truncated CCT Domain Gene in albostrians Barley. The Plant Cell, 31(7), 1430–1445. https://doi.org/10.1105/tpc.19.00132

Li, Y., Mukherjee, I., Thum, K. E., Tanurdzic, M., Katari, M. S., Obertello, M., Edwards, M. B., McCombie, W. R., Martienssen, R. A., & Coruzzi, G. M. (2015). The histone methyltransferase SDG8 mediates the epigenetic modification of light and carbon responsive genes in plants. Genome Biology, 16(1), 79. https://doi.org/10.1186/s13059-015-0640-2

Liebers, M., Cozzi, C., Uecker, F., Chambon, L., Blanvillain, R., & Pfannschmidt, T. (2022). Biogenic signals from plastids and their role in chloroplast development. Journal of Experimental Botany, 73, 7105. https://doi.org/10.1093/jxb/erac344

Liebers, M., Gillet, F.-X., Israel, A., Pounot, K., Chambon, L., Chieb, M., Chevalier, F., Ruedas, R., Favier, A., Gans, P., Boeri Erba, E., Cobessi, D., Pfannschmidt, T., & Blanvillain, R. (2020). Nucleo-plastidic PAP8/pTAC6 couples chloroplast formation with photomorphogenesis. The EMBO Journal, 39(22), e104941. https://doi.org/10.15252/embj.2020104941

Liebers, M., Grübler, B., Chevalier, F., Lerbs-Mache, S., Merendino, L., Blanvillain, R., & Pfannschmidt, T. (2017). Regulatory Shifts in Plastid Transcription Play a Key Role in Morphological Conversions of Plastids during Plant Development. Frontiers in Plant Science, 8, 23. https://doi.org/10.3389/fpls.2017.00023

Littlejohn, G. R., Breen, S., Smirnoff, N., & Grant, M. (2021). Chloroplast immunity illuminated. New Phytologist, 229(6), 3088–3107. https://doi.org/10.1111/nph.17076

Liu, J.-X., & Howell, S. H. (2016). Managing the protein folding demands in the endoplasmic reticulum of plants. New Phytologist, 211(2), 418–428. https://doi.org/10.1111/nph.13915

Liu, L., McNeilage, R. T., Shi, L.-X., & Theg, S. M. (2014). ATP requirement for chloroplast protein import is set by the Km for ATP hydrolysis of stromal Hsp70 in Physcomitrella patens. The Plant Cell, 26(3), 1246–1255. https://doi.org/10.1105/tpc.113.121822

Liu, Z.-W., Simmons, C. H., & Zhong, X. (2022). Linking transcriptional silencing with chromatin remodeling, folding, and positioning in the nucleus. Current Opinion in Plant Biology, 69, 102261. https://doi.org/10.1016/j.pbi.2022.102261

Long, J. A., Ohno, C., Smith, Z. R., & Meyerowitz, E. M. (2006). TOPLESS Regulates Apical Embryonic Fate in Arabidopsis. Science, 312(5779), 1520–1523. https://doi.org/10.1126/science.1123841

Lopez-Juez, E., & Pyke, K. A. (2005). Plastids unleashed: Their development and their integration in plant development. The International Journal of Developmental Biology, 49(5–6), Article 5–6. https://doi.org/10.1387/ijdb.051997el

Lu, F., Cui, X., Zhang, S., Jenuwein, T., & Cao, X. (2011). Arabidopsis REF6 is a histone H3 lysine 27 demethylase. Nature Genetics, 43(7), 715–719. https://doi.org/10.1038/ng.854

Ma, R., Sun, L., Chen, X., Mei, B., Chang, G., Wang, M., & Zhao, D. (2016). Proteomic Analyses Provide Novel Insights into Plant Growth and Ginsenoside Biosynthesis in Forest Cultivated Panax ginseng (F. Ginseng). Frontiers in Plant Science, 7, 1. https://doi.org/10.3389/fpls.2016.00001

Mamaeva, A., Taliansky, M., Filippova, A., Love, A. J., Golub, N., & Fesenko, I. (2020). The role of chloroplast protein remodeling in stress responses and shaping of the plant peptidome. New Phytologist, 227(5), 1326–1334. https://doi.org/10.1111/nph.16620

Mao, C., He, J., Liu, L., Deng, Q., Yao, X., Liu, C., Qiao, Y., Li, P., & Ming, F. (2020). OsNAC2 integrates auxin and cytokinin pathways to modulate rice root development. Plant Biotechnology Journal, 18(2), 429–442. https://doi.org/10.1111/pbi.13209

Marasca, F., Bodega, B., & Orlando, V. (2018). How Polycomb-Mediated Cell Memory Deals With a Changing Environment. BioEssays, 40(4), 1700137. https://doi.org/10.1002/bies.201700137

Margueron, R., & Reinberg, D. (2011). The Polycomb complex PRC2 and its mark in life. Nature, 469(7330), 343–349. https://doi.org/10.1038/nature09784

Margulis, L. (1975). Symbiotic theory of the origin of eukaryotic organelles; criteria for proof. Symposia of the Society for Experimental Biology, 29, 21–38.

Martín, G., Leivar, P., Ludevid, D., Tepperman, J. M., Quail, P. H., & Monte, E. (2016). Phytochrome and retrograde signalling pathways converge to antagonistically regulate a light-induced transcriptional network. Nature Communications, 7, 11431. https://doi.org/10.1038/ncomms11431

Martin, W., & Herrmann, R. G. (1998). Gene transfer from organelles to the nucleus: How much, what happens, and Why? Plant Physiology, 118(1), 9–17. https://doi.org/10.1104/pp.118.1.9

Martin, W., Stoebe, B., Goremykin, V., Hapsmann, S., Hasegawa, M., & Kowallik, K. V. (1998). Gene transfer to the nucleus and the evolution of chloroplasts. Nature, 393(6681), 162–165. https://doi.org/10.1038/30234

Melonek, J., Oetke, S., & Krupinska, K. (2016). Multifunctionality of plastid nucleoids as revealed by proteome analyses. Biochimica et Biophysica Acta (BBA) - Proteins and Proteomics, 1864(8), 1016–1038. https://doi.org/10.1016/j.bbapap.2016.03.009

Missihoun, T. D., Kirch, H.-H., & Bartels, D. (2012). T-DNA insertion mutants reveal complex expression patterns of the aldehyde dehydrogenase 3H1 locus in Arabidopsis thaliana. Journal of Experimental Botany, 63(10), 3887–3898. https://doi.org/10.1093/jxb/ers081

Mochizuki, N., Brusslan, J. A., Larkin, R., Nagatani, A., & Chory, J. (2001). Arabidopsis genomes uncoupled 5 (GUN5) mutant reveals the involvement of Mg-chelatase H subunit in plastid-to-nucleus signal transduction. Proceedings of the National Academy of Sciences, 98(4), 2053–2058. https://doi.org/10.1073/pnas.98.4.2053

Mochizuki, N., Susek, R., & Chory, J. (1996). An Intracellular Signal Transduction Pathway between the Chloroplast and Nucleus Is Involved in De-Etiolation. Plant Physiology, 112(4), 1465–1469. https://doi.org/10.1104/pp.112.4.1465

Morera, L., Lübbert, M., & Jung, M. (2016). Targeting histone methyltransferases and demethylases in clinical trials for cancer therapy. Clinical Epigenetics, 8(1), 57. https://doi.org/10.1186/s13148-016-0223-4

Mozgova, I., & Hennig, L. (2015). The Polycomb Group Protein Regulatory Network. Annual Review of Plant Biology, 66(1), 269–296. https://doi.org/10.1146/annurev-arplant-043014-115627

Mulo, P., Pursiheimo, S., Hou, C.-X., Tyystjärvi, T., & Aro, E.-M. (2003). Multiple effects of antibiotics on chloroplast and nuclear gene expression. Functional Plant Biology: FPB, 30(11), 1097–1103. https://doi.org/10.1071/FP03149

Neuhaus, H. E., & Emes, M. J. (2000). NONPHOTOSYNTHETIC METABOLISM IN PLASTIDS. Annual Review of Plant Physiology and Plant Molecular Biology, 51, 111–140. https://doi.org/10.1146/annurev.arplant.51.1.111

Nevarez, P. A., Qiu, Y., Inoue, H., Yoo, C. Y., Benfey, P. N., Schnell, D. J., & Chen, M. (2017). Mechanism of Dual Targeting of the Phytochrome Signaling Component HEMERA/pTAC12 to Plastids and the Nucleus. Plant Physiology, 173(4), 1953–1966. https://doi.org/10.1104/pp.16.00116

Ning, Y.-Q., Chen, Q., Lin, R.-N., Li, Y.-Q., Li, L., Chen, S., & He, X.-J. (2019). The HDA19 histone deacetylase complex is involved in the regulation of flowering time in a photoperiod-dependent manner. The Plant Journal, 98(3), 448–464. https://doi.org/10.1111/tpj.14229

Noordally, Z. B., Ishii, K., Atkins, K. A., Wetherill, S. J., Kusakina, J., Walton, E. J., Kato, M., Azuma, M., Tanaka, K., Hanaoka, M., & Dodd, A. N. (2013). Circadian Control of Chloroplast Transcription by a Nuclear-Encoded Timing Signal. Science, 339(6125), 1316–1319. https://doi.org/10.1126/science.1230397

Norén, L., Kindgren, P., Stachula, P., Rühl, M., Eriksson, M. E., Hurry, V., & Strand, Å. (2016). Circadian and Plastid Signaling Pathways Are Integrated to Ensure Correct Expression of the CBF and COR Genes during Photoperiodic Growth. Plant Physiology, 171(2), 1392–1406. https://doi.org/10.1104/pp.16.00374

Oelmüller, R., Levitan, I., Bergfeld, R., Rajasekhar, V. K., & Mohr, H. (1986). Expression of nuclear genes as affected by treatments acting on the plastids. Planta, 168(4), 482–492. https://doi.org/10.1007/BF00392267

Oh, E., Zhu, J.-Y., & Wang, Z.-Y. (2012). Interaction between BZR1 and PIF4 integrates brassinosteroid and environmental responses. Nature Cell Biology, 14(8), Article 8. https://doi.org/10.1038/ncb2545

Page, M. T., Garcia-Becerra, T., Smith, A. G., & Terry, M. J. (2020). Overexpression of chloroplast-targeted ferrochelatase 1 results in a genomes uncoupled chloroplast-to-nucleus retrograde signalling phenotype. Philosophical Transactions of the Royal Society B: Biological Sciences, 375(1801), 20190401. https://doi.org/10.1098/rstb.2019.0401

Page, M. T., McCormac, A. C., Smith, A. G., & Terry, M. J. (2017). Singlet oxygen initiates a plastid signal controlling photosynthetic gene expression. New Phytologist, 213(3), 1168–1180. https://doi.org/10.1111/nph.14223

Paieri, F., Tadini, L., Manavski, N., Kleine, T., Ferrari, R., Morandini, P., Pesaresi, P., Meurer, J., & Leister, D. (2018). The DEAD-box RNA Helicase RH50 Is a 23S-4.5S rRNA Maturation Factor that Functionally Overlaps with the Plastid Signaling Factor GUN1. Plant Physiology, 176(1), 634–648. https://doi.org/10.1104/pp.17.01545

Pandey, R., Müller, A., Napoli, C. A., Selinger, D. A., Pikaard, C. S., Richards, E. J., Bender, J., Mount, D. W., & Jorgensen, R. A. (2002). Analysis of histone acetyltransferase and histone deacetylase families of Arabidopsis thaliana suggests functional diversification of chromatin modification among multicellular eukaryotes. Nucleic Acids Research, 30(23), 5036–5055. https://doi.org/10.1093/nar/gkf660

Parihar, V., Arya, D., Walia, A., Tyagi, V., Dangwal, M., Verma, V., Khurana, R., Boora, N., Kapoor, S., & Kapoor, M. (2019). Functional characterization of LIKE HETEROCHROMATIN PROTEIN 1 in the moss Physcomitrella patens: Its conserved protein interactions in land plants. The Plant Journal, 97(2), 221–239. https://doi.org/10.1111/tpj.14182

Parks, B. M., & Quail, P. H. (1991). Phytochrome-Deficient hy1 and hy2 Long Hypocotyl Mutants of Arabidopsis Are Defective in Phytochrome Chromophore Biosynthesis. The Plant Cell, 3(11), 1177–1186. https://doi.org/10.2307/3869225

Peng, M., Ying, P., Liu, X., Li, C., Xia, R., Li, J., & Zhao, M. (2017). Genome-Wide Identification of Histone Modifiers and Their Expression Patterns during Fruit Abscission in Litchi. Frontiers in Plant Science, 8, 639. https://doi.org/10.3389/fpls.2017.00639

Perrella, G., Lopez-Vernaza, M. A., Carr, C., Sani, E., Gosselé, V., Verduyn, C., Kellermeier, F., Hannah, M. A., & Amtmann, A. (2013). Histone deacetylase complex1 expression level titrates plant growth and abscisic acid sensitivity in Arabidopsis. The Plant Cell, 25(9), 3491–3505. https://doi.org/10.1105/tpc.113.114835

Pesaresi, P., & Kim, C. (2019). Current understanding of GUN1: A key mediator involved in biogenic retrograde signaling. Plant Cell Reports, 38(7), 819–823. https://doi.org/10.1007/s00299-019-02383-4

Petrillo, E., Godoy Herz, M. A., Fuchs, A., Reifer, D., Fuller, J., Yanovsky, M. J., Simpson, C., Brown, J. W. S., Barta, A., Kalyna, M., & Kornblihtt, A. R. (2014). A Chloroplast Retrograde Signal Regulates Nuclear Alternative Splicing. Science, 344(6182), 427–430. https://doi.org/10.1126/science.1250322

Pikaard, C. S., & Mittelsten Scheid, O. (2014). Epigenetic Regulation in Plants. Cold Spring Harbor Perspectives in Biology, 6(12), a019315. https://doi.org/10.1101/cshperspect.a019315

Pipitone, R., Eicke, S., Pfister, B., Glauser, G., Falconet, D., Uwizeye, C., Pralon, T., Zeeman, S. C., Kessler, F., & Demarsy, E. (n.d.). A multifaceted analysis reveals two distinct phases of chloroplast biogenesis during de-etiolation in Arabidopsis. eLife, 10, e62709. https://doi.org/10.7554/eLife.62709

Plastid-produced interorgannellar stress signal MEcPP potentiates induction of the unfolded protein response in endoplasmic reticulum | PNAS. (n.d.). Retrieved 20 August 2023, from https://www.pnas.org/doi/full/10.1073/pnas.1504828112

Pogson, B. J., Ganguly, D., & Albrecht-Borth, V. (2015). Insights into chloroplast biogenesis and development. Biochimica et Biophysica Acta (BBA) - Bioenergetics, 1847(9), 1017–1024. https://doi.org/10.1016/j.bbabio.2015.02.003

Ponce-Toledo, R. I., López-García, P., & Moreira, D. (2019). Horizontal and endosymbiotic gene transfer in early plastid evolution. The New Phytologist, 224(2), 618–624. https://doi.org/10.1111/nph.15965

Probst, A. V., & Mittelsten Scheid, O. (2015). Stress-induced structural changes in plant chromatin. Current Opinion in Plant Biology, 27, 8–16. https://doi.org/10.1016/j.pbi.2015.05.011

Pu, L., Liu, M.-S., Kim, S. Y., Chen, L.-F. O., Fletcher, J. C., & Sung, Z. R. (2013). EMBRYONIC FLOWER1 and ULTRAPETALA1 Act Antagonistically on Arabidopsis Development and Stress Response1[W]. Plant Physiology, 162(2), 812–830. https://doi.org/10.1104/pp.112.213223

Puranik, S., Sahu, P. P., Srivastava, P. S., & Prasad, M. (2012). NAC proteins: Regulation and role in stress tolerance. Trends in Plant Science, 17(6), 369–381. https://doi.org/10.1016/j.tplants.2012.02.004

Pyke, K. (2007). Plastid biogenesis and differentiation. In R. Bock (Ed.), Cell and Molecular Biology of Plastids (pp. 1–28). Springer. https://doi.org/10.1007/4735_2007_0226

Ramirez-Prado, J. S., Latrasse, D., Rodriguez-Granados, N. Y., Huang, Y., Manza-Mianza, D., Brik-Chaouche, R., Jaouannet, M., Citerne, S., Bendahmane, A., Hirt, H., Raynaud, C., & Benhamed, M. (2019). The Polycomb protein LHP1 regulates Arabidopsis thaliana stress responses through the repression of the MYC2-dependent branch of immunity. The Plant Journal: For Cell and Molecular Biology, 100(6), 1118–1131. https://doi.org/10.1111/tpj.14502

Richardson, L. G. L., & Schnell, D. J. (2020). Origins, function, and regulation of the TOC–TIC general protein import machinery of plastids. Journal of Experimental Botany, 71(4), 1226–1238. https://doi.org/10.1093/jxb/erz517

Richly, E., Dietzmann, A., Biehl, A., Kurth, J., Laloi, C., Apel, K., Salamini, F., & Leister, D. (2003). Covariations in the nuclear chloroplast transcriptome reveal a regulatory master-switch. EMBO Reports, 4(5), 491–498. https://doi.org/10.1038/sj.embor.embor828

Richter, A. S., Nägele, T., Grimm, B., Kaufmann, K., Schroda, M., Leister, D., & Kleine, T. (2023). Retrograde signaling in plants: A critical review focusing on the GUN pathway and beyond. Plant Communications, 4(1), 100511. https://doi.org/10.1016/j.xplc.2022.100511

Richter, R., Behringer, C., Müller, I. K., & Schwechheimer, C. (2010). The GATA-type transcription factors GNC and GNL/CGA1 repress gibberellin signaling downstream from DELLA proteins and PHYTOCHROME-INTERACTING FACTORS. Genes & Development, 24(18), 2093–2104. https://doi.org/10.1101/gad.594910

Richter, R., Behringer, C., Zourelidou, M., & Schwechheimer, C. (2013). Convergence of auxin and gibberellin signaling on the regulation of the GATA transcription factors GNC and GNL in Arabidopsis thaliana. Proceedings of the National Academy of Sciences of the United States of America, 110(32), 13192–13197. https://doi.org/10.1073/pnas.1304250110

Rossini, L., Cribb, L., Martin, D. J., & Langdale, J. A. (2001). The maize golden2 gene defines a novel class of transcriptional regulators in plants. The Plant Cell, 13(5), 1231–1244. https://doi.org/10.1105/tpc.13.5.1231

Ruckle, M. E., Burgoon, L. D., Lawrence, L. A., Sinkler, C. A., & Larkin, R. M. (2012). Plastids Are Major Regulators of Light Signaling in Arabidopsis. Plant Physiology, 159(1), 366–390. https://doi.org/10.1104/pp.112.193599

Ruckle, M. E., DeMarco, S. M., & Larkin, R. M. (2007). Plastid Signals Remodel Light Signaling Networks and Are Essential for Efficient Chloroplast Biogenesis in Arabidopsis. The Plant Cell, 19(12), 3944–3960. https://doi.org/10.1105/tpc.107.054312

Ruckle, M. E., & Larkin, R. M. (2009). Plastid signals that affect photomorphogenesis in Arabidopsis thaliana are dependent on GENOMES UNCOUPLED 1 and cryptochrome 1. New Phytologist, 182(2), 367–379. https://doi.org/10.1111/j.1469-8137.2008.02729.x

Rutowicz, K., Lirski, M., Mermaz, B., Teano, G., Schubert, J., Mestiri, I., Kroteń, M. A., Fabrice, T. N., Fritz, S., Grob, S., Ringli, C., Cherkezyan, L., Barneche, F., Jerzmanowski, A., & Baroux, C. (2019).

Linker histones are fine-scale chromatin architects modulating developmental decisions in Arabidopsis. Genome Biology, 20(1), 157. https://doi.org/10.1186/s13059-019-1767-3

Saha, A., Wittmeyer, J., & Cairns, B. R. (2006). Chromatin remodelling: The industrial revolution of DNA around histones. Nature Reviews Molecular Cell Biology, 7(6), Article 6. https://doi.org/10.1038/nrm1945

Sahu, P. P., Pandey, G., Sharma, N., Puranik, S., Muthamilarasan, M., & Prasad, M. (2013). Epigenetic mechanisms of plant stress responses and adaptation. Plant Cell Reports, 32(8), 1151–1159. https://doi.org/10.1007/s00299-013-1462-x

Sakuraba, Y., Kim, D., Han, S.-H., Kim, S.-H., Piao, W., Yanagisawa, S., An, G., & Paek, N.-C. (2020). Multilayered Regulation of Membrane-Bound ONAC054 Is Essential for Abscisic Acid-Induced Leaf Senescence in Rice. The Plant Cell, 32(3), 630–649. https://doi.org/10.1105/tpc.19.00569

Samo, N., Ebert, A., Kopka, J., & Mozgová, I. (2021). Plant chromatin, metabolism and development – an intricate crosstalk. Current Opinion in Plant Biology, 61, 102002. https://doi.org/10.1016/j.pbi.2021.102002

Serrano, I., Audran, C., & Rivas, S. (2016). Chloroplasts at work during plant innate immunity. Journal of Experimental Botany, 67(13), 3845–3854. https://doi.org/10.1093/jxb/erw088

Servet, C., Conde e Silva, N., & Zhou, D.-X. (2010). Histone Acetyltransferase AtGCN5/HAG1 Is a Versatile Regulator of Developmental and Inducible Gene Expression in Arabidopsis. Molecular Plant, 3(4), 670–677. https://doi.org/10.1093/mp/ssq018

Shi, C., Wang, S., Xia, E.-H., Jiang, J.-J., Zeng, F.-C., & Gao, L.-Z. (2016). Full transcription of the chloroplast genome in photosynthetic eukaryotes. Scientific Reports, 6(1), Article 1. https://doi.org/10.1038/srep30135

Shimizu, T., Kacprzak, S. M., Mochizuki, N., Nagatani, A., Watanabe, S., Shimada, T., Tanaka, K., Hayashi, Y., Arai, M., Leister, D., Okamoto, H., Terry, M. J., & Masuda, T. (2019). The retrograde signaling protein GUN1 regulates tetrapyrrole biosynthesis. Proceedings of the National Academy of Sciences, 116(49), 24900–24906. https://doi.org/10.1073/pnas.1911251116

Simon, J. A., & Kingston, R. E. (2009). Mechanisms of polycomb gene silencing: Knowns and unknowns. Nature Reviews. Molecular Cell Biology, 10(10), 697–708. https://doi.org/10.1038/nrm2763

Simonini, S., Bemer, M., Bencivenga, S., Gagliardini, V., Pires, N. D., Desvoyes, B., Van Der Graaff, E., Gutierrez, C., & Grossniklaus, U. (2021). The Polycomb group protein MEDEA controls cell proliferation and embryonic patterning in Arabidopsis. Developmental Cell, 56(13), 1945-1960.e7. https://doi.org/10.1016/j.devcel.2021.06.004

Sjuts, I., Soll, J., & Bölter, B. (2017). Import of Soluble Proteins into Chloroplasts and Potential Regulatory Mechanisms. Frontiers in Plant Science, 8. https://www.frontiersin.org/articles/10.3389/fpls.2017.00168

Smale, S. T., Tarakhovsky, A., & Natoli, G. (2014). Chromatin contributions to the regulation of innate immunity. Annual Review of Immunology, 32, 489–511. https://doi.org/10.1146/annurev-immunol-031210-101303

Solymosi, K., & Schoefs, B. (2010). Etioplast and etio-chloroplast formation under natural conditions: The dark side of chlorophyll biosynthesis in angiosperms. Photosynthesis Research, 105(2), 143–166. https://doi.org/10.1007/s11120-010-9568-2

Song, Y., Feng, L., Alyafei, M. A. M., Jaleel, A., & Ren, M. (2021). Function of Chloroplasts in Plant Stress Responses. International Journal of Molecular Sciences, 22(24), 13464. https://doi.org/10.3390/ijms222413464

Spetea, C., Rintamäki, E., & Schoefs, B. (2014). Changing the light environment: Chloroplast signalling and response mechanisms. Philosophical Transactions of the Royal Society B: Biological Sciences, 369(1640), 20130220. https://doi.org/10.1098/rstb.2013.0220

Stadnichuk, I. N., & Kusnetsov, V. V. (2021). Endosymbiotic Origin of Chloroplasts in Plant Cells' Evolution. Russian Journal of Plant Physiology, 68(1), 1–16. https://doi.org/10.1134/S1021443721010179

Stenbaek, A., Hansson, A., Wulff, R. P., Hansson, M., Dietz, K.-J., & Jensen, P. E. (2008). NADPH-dependent thioredoxin reductase and 2-Cys peroxiredoxins are needed for the protection of Mg-protoporphyrin monomethyl ester cyclase. FEBS Letters, 582(18), 2773–2778. https://doi.org/10.1016/j.febslet.2008.07.006

Stiti, N., Missihoun, T., Kotchoni, S., Kirch, H.-H., & Bartels, D. (2011). Aldehyde Dehydrogenases in Arabidopsis thaliana: Biochemical Requirements, Metabolic Pathways, and Functional Analysis. Frontiers in Plant Science, 2. https://www.frontiersin.org/articles/10.3389/fpls.2011.00065

Strand, A., Asami, T., Alonso, J., Ecker, J. R., & Chory, J. (2003). Chloroplast to nucleus communication triggered by accumulation of Mg-protoporphyrinIX. Nature, 421(6918), 79–83. https://doi.org/10.1038/nature01204

Su, P.-H., & Li, H. (2010). Stromal Hsp70 is important for protein translocation into pea and Arabidopsis chloroplasts. The Plant Cell, 22(5), 1516–1531. https://doi.org/10.1105/tpc.109.071415

Su, Y., Wang, S., Zhang, F., Zheng, H., Liu, Y., Huang, T., & Ding, Y. (2017). Phosphorylation of Histone H2A at Serine 95: A Plant-Specific Mark Involved in Flowering Time Regulation and H2A.Z Deposition. The Plant Cell, 29(9), 2197–2213. https://doi.org/10.1105/tpc.17.00266

Sun, Y., & Zerges, W. (2015). Translational regulation in chloroplasts for development and homeostasis. Biochimica et Biophysica Acta (BBA) - Bioenergetics, 1847(9), 809–820. https://doi.org/10.1016/j.bbabio.2015.05.008

Susek, R. E., Ausubel, F. M., & Chory, J. (1993). Signal transduction mutants of arabidopsis uncouple nuclear CAB and RBCS gene expression from chloroplast development. Cell, 74(5), 787–799. https://doi.org/10.1016/0092-8674(93)90459-4

Szymańska, R., Ślesak, I., Orzechowska, A., & Kruk, J. (2017). Physiological and biochemical responses to high light and temperature stress in plants. Environmental and Experimental Botany, 139, 165–177. https://doi.org/10.1016/j.envexpbot.2017.05.002

Tabrizi, S. T., Sawicki, A., Zhou, S., Luo, M., & Willows, R. D. (2016). GUN4-Protoporphyrin IX Is a Singlet Oxygen Generator with Consequences for Plastid Retrograde Signaling *. Journal of Biological Chemistry, 291(17), 8978–8984. https://doi.org/10.1074/jbc.C116.719989

Tadini, L., Peracchio, C., Trotta, A., Colombo, M., Mancini, I., Jeran, N., Costa, A., Faoro, F., Marsoni, M., Vannini, C., Aro, E.-M., & Pesaresi, P. (2020). GUN1 influences the accumulation of NEP-dependent transcripts and chloroplast protein import in Arabidopsis cotyledons upon perturbation of chloroplast protein homeostasis. The Plant Journal, 101(5), 1198–1220. https://doi.org/10.1111/tpj.14585

Tadini, L., Pesaresi, P., Kleine, T., Rossi, F., Guljamow, A., Sommer, F., Mühlhaus, T., Schroda, M., Masiero, S., Pribil, M., Rothbart, M., Hedtke, B., Grimm, B., & Leister, D. (2016). GUN1 Controls Accumulation of the Plastid Ribosomal Protein S1 at the Protein Level and Interacts with Proteins Involved in Plastid Protein Homeostasis. Plant Physiology, 170(3), 1817–1830. https://doi.org/10.1104/pp.15.02033

Tanaka, M., Kikuchi, A., & Kamada, H. (2008). The Arabidopsis histone deacetylases HDA6 and HDA19 contribute to the repression of embryonic properties after germination. Plant Physiology, 146(1), 149–161. https://doi.org/10.1104/pp.107.111674

Tanaka, R., Kobayashi, K., & Masuda, T. (2011). Tetrapyrrole Metabolism in Arabidopsis thaliana. The Arabidopsis Book, 2011(9). https://doi.org/10.1199/tab.0145

Ueda, M., & Seki, M. (2020). Histone Modifications Form Epigenetic Regulatory Networks to Regulate Abiotic Stress Response1 [OPEN]. Plant Physiology, 182(1), 15–26. https://doi.org/10.1104/pp.19.00988

Van Dingenen, J., De Milde, L., Vermeersch, M., Maleux, K., De Rycke, R., De Bruyne, M., Storme, V., Gonzalez, N., Dhondt, S., & Inzé, D. (2016). Chloroplasts Are Central Players in Sugar-Induced Leaf Growth1[OPEN]. Plant Physiology, 171(1), 590–605. https://doi.org/10.1104/pp.15.01669

Vlachonasios, K. E., Thomashow, M. F., & Triezenberg, S. J. (2003). Disruption Mutations of ADA2b and GCN5 Transcriptional Adaptor Genes Dramatically Affect Arabidopsis Growth, Development, and Gene Expression[W]. The Plant Cell, 15(3), 626–638. https://doi.org/10.1105/tpc.007922

Walley, J., Xiao, Y., Wang, J.-Z., Baidoo, E. E., Keasling, J. D., Shen, Z., Briggs, S. P., & Dehesh, K. (2015). Plastid-produced interorgannellar stress signal MEcPP potentiates induction of the unfolded protein response in endoplasmic reticulum. Proceedings of the National Academy of Sciences, 112(19), 6212–6217. https://doi.org/10.1073/pnas.1504828112

Wang, P., Khoshravesh, R., Karki, S., Tapia, R., Balahadia, C. P., Bandyopadhyay, A., Quick, W. P., Furbank, R., Sage, T. L., & Langdale, J. A. (2017). Re-creation of a Key Step in the Evolutionary Switch from C3 to C4 Leaf Anatomy. Current Biology: CB, 27(21), 3278-3287.e6. https://doi.org/10.1016/j.cub.2017.09.040

Wang, X., Chen, J., Xie, Z., Liu, S., Nolan, T., Ye, H., Zhang, M., Guo, H., Schnable, P. S., Li, Z., & Yin, Y. (2014). Histone Lysine Methyltransferase SDG8 Is Involved in Brassinosteroid-Regulated Gene Expression in Arabidopsis thaliana. Molecular Plant, 7(8), 1303–1315. https://doi.org/10.1093/mp/ssu056

Waters, M. T., & Langdale, J. A. (2009). The making of a chloroplast. The EMBO Journal, 28(19), 2861–2873. https://doi.org/10.1038/emboj.2009.264

Waters, M. T., Wang, P., Korkaric, M., Capper, R. G., Saunders, N. J., & Langdale, J. A. (2009). GLK transcription factors coordinate expression of the photosynthetic apparatus in Arabidopsis. The Plant Cell, 21(4), 1109–1128. https://doi.org/10.1105/tpc.108.065250

Witte, C.-P., & Herde, M. (2020). Nucleotide Metabolism in Plants1 [OPEN]. Plant Physiology, 182(1), 63–78. https://doi.org/10.1104/pp.19.00955

Woodson, J. D., & Chory, J. (2008). Coordination of gene expression between organellar and nuclear genomes. Nature Reviews. Genetics, 9(5), 383–395. https://doi.org/10.1038/nrg2348

Woodson, J. D., Perez-Ruiz, J. M., & Chory, J. (2011). Heme synthesis by plastid ferrochelatase I regulates nuclear gene expression in plants. Current Biology: CB, 21(10), 897–903. https://doi.org/10.1016/j.cub.2011.04.004

Woodson, J. D., Perez-Ruiz, J. M., Schmitz, R. J., Ecker, J. R., & Chory, J. (2013). Sigma factor-mediated plastid retrograde signals control nuclear gene expression. The Plant Journal : For Cell and Molecular Biology, 73(1), 1–13. https://doi.org/10.1111/tpj.12011

Wu, G.-Z., & Bock, R. (2021). GUN control in retrograde signaling: How GENOMES UNCOUPLED proteins adjust nuclear gene expression to plastid biogenesis. The Plant Cell, 33(3), 457–474. https://doi.org/10.1093/plcell/koaa048

Wu, G.-Z., Chalvin, C., Hoelscher, M., Meyer, E. H., Wu, X. N., & Bock, R. (2018). Control of Retrograde Signaling by Rapid Turnover of GENOMES UNCOUPLED11[OPEN]. Plant Physiology, 176(3), 2472–2495. https://doi.org/10.1104/pp.18.00009

Wu, G.-Z., Meyer, E. H., Richter, A. S., Schuster, M., Ling, Q., Schöttler, M. A., Walther, D., Zoschke, R., Grimm, B., Jarvis, R. P., & Bock, R. (2019). Control of retrograde signalling by protein import and cytosolic folding stress. Nature Plants, 5(5), 525–538. https://doi.org/10.1038/s41477-019-0415-y

Wu, G.-Z., Meyer, E. H., Wu, S., & Bock, R. (2019). Extensive Posttranscriptional Regulation of Nuclear Gene Expression by Plastid Retrograde Signals. Plant Physiology, 180(4), 2034–2048. https://doi.org/10.1104/pp.19.00421

Xiao, J., Jin, R., Yu, X., Shen, M., Wagner, J. D., Pai, A., Song, C., Zhuang, M., Klasfeld, S., He, C., Santos, A. M., Helliwell, C., Pruneda-Paz, J. L., Kay, S. A., Lin, X., Cui, S., Garcia, M. F., Clarenz, O., Goodrich, J., … Wagner, D. (2017). Cis and trans determinants of epigenetic silencing by Polycomb repressive complex 2 in Arabidopsis. Nature Genetics, 49(10), Article 10. https://doi.org/10.1038/ng.3937

Xiao, Y., Savchenko, T., Baidoo, E. E. K., Chehab, W. E., Hayden, D. M., Tolstikov, V., Corwin, J. A., Kliebenstein, D. J., Keasling, J. D., & Dehesh, K. (2012). Retrograde Signaling by the Plastidial Metabolite MEcPP Regulates Expression of Nuclear Stress-Response Genes. Cell, 149(7), 1525–1535. https://doi.org/10.1016/j.cell.2012.04.038

Xu, X., Chi, W., Sun, X., Feng, P., Guo, H., Li, J., Lin, R., Lu, C., Wang, H., Leister, D., & Zhang, L. (2016). Convergence of light and chloroplast signals for de-etiolation through ABI4–HY5 and COP1. Nature Plants, 2(6), Article 6. https://doi.org/10.1038/nplants.2016.66

Yamaguchi, N. (2021). Removal of H3K27me3 by JMJ Proteins Controls Plant Development and Environmental Responses in Arabidopsis. Frontiers in Plant Science, 12, 687416. https://doi.org/10.3389/fpls.2021.687416

Yoon, H. S., Hackett, J. D., Ciniglia, C., Pinto, G., & Bhattacharya, D. (2004). A Molecular Timeline for the Origin of Photosynthetic Eukaryotes. Molecular Biology and Evolution, 21(5), 809–818. https://doi.org/10.1093/molbev/msh075

Yu, C.-W., Liu, X., Luo, M., Chen, C., Lin, X., Tian, G., Lu, Q., Cui, Y., & Wu, K. (2011). HISTONE DEACETYLASE6 Interacts with FLOWERING LOCUS D and Regulates Flowering in Arabidopsis. Plant Physiology, 156(1), 173–184. https://doi.org/10.1104/pp.111.174417

Yuan, Y., Xu, X., Gong, Z., Tang, Y., Wu, M., Yan, F., Zhang, X., Zhang, Q., Yang, F., Hu, X., Yang, Q., Luo, Y., Mei, L., Zhang, W., Jiang, C.-Z., Lu, W., Li, Z., & Deng, W. (2019). Auxin response factor 6A regulates photosynthesis, sugar accumulation, and fruit development in tomato. Horticulture Research, 6, 85. https://doi.org/10.1038/s41438-019-0167-x

Žádníková, P., Smet, D., Zhu, Q., Straeten, D. V. D., & Benková, E. (2015). Strategies of seedlings to overcome their sessile nature: Auxin in mobility control. Frontiers in Plant Science, 6, 218. https://doi.org/10.3389/fpls.2015.00218

Zhang, J., Ruhlman, T. A., Sabir, J., Blazier, J. C., & Jansen, R. K. (2015). Coordinated Rates of Evolution between Interacting Plastid and Nuclear Genes in Geraniaceae. The Plant Cell, 27(3), 563–573. https://doi.org/10.1105/tpc.114.134353

Zhang, Y., Zhang, A., Li, X., & Lu, C. (2020). The Role of Chloroplast Gene Expression in Plant Responses to Environmental Stress. International Journal of Molecular Sciences, 21(17), Article 17. https://doi.org/10.3390/ijms21176082

Zhang, Z.-W., Zhang, G.-C., Zhu, F., Zhang, D.-W., & Yuan, S. (2015). The roles of tetrapyrroles in plastid retrograde signaling and tolerance to environmental stresses. Planta, 242(6), 1263–1276. https://doi.org/10.1007/s00425-015-2384-3

Zhao, S., Zhang, B., Yang, M., Zhu, J., & Li, H. (2018). Systematic Profiling of Histone Readers in Arabidopsis thaliana. Cell Reports, 22(4), 1090–1102. https://doi.org/10.1016/j.celrep.2017.12.099

Zheng, S., Hu, H., Ren, H., Yang, Z., Qiu, Q., Qi, W., Liu, X., Chen, X., Cui, X., Li, S., Zhou, B., Sun, D., Cao, X., & Du, J. (2019). The Arabidopsis H3K27me3 demethylase JUMONJI 13 is a temperature and photoperiod dependent flowering repressor. Nature Communications, 10, 1303. https://doi.org/10.1038/s41467-019-09310-x

Zhu, Y., Klasfeld, S., Jeong, C. W., Jin, R., Goto, K., Yamaguchi, N., & Wagner, D. (2020). TERMINAL FLOWER 1-FD complex target genes and competition with FLOWERING LOCUS T. Nature Communications, 11, 5118. https://doi.org/10.1038/s41467-020-18782-1

Zidovska, A. (2020). The rich inner life of the cell nucleus: Dynamic organization, active flows, and emergent rheology. Biophysical Reviews, 12(5), 1093–1106. https://doi.org/10.1007/s12551-020-00761-x

Zubo, Y. O., Blakley, I. C., Franco-Zorrilla, J. M., Yamburenko, M. V., Solano, R., Kieber, J. J., Loraine, A. E., & Schaller, G. E. (2018). Coordination of Chloroplast Development through the Action of the GNC and GLK Transcription Factor Families. Plant Physiology, 178(1), 130–147. https://doi.org/10.1104/pp.18.00414