

Chapter 18

Intracellular Signaling and Chlorophyll Synthesis

Robert M. Larkin*

*MSU-DOE Plant Research Laboratory and Department of Biochemistry and Molecular Biology,
Michigan State University, East Lansing, MI 48824, USA*

Summary	289
I. Introduction	289
II. Chlorophyll Biosynthetic Mutant, Inhibitor, and Feeding Studies	290
III. Plastid-to-Nucleus Signaling Mutants Inhibit Mg-Porphyrin Accumulation	293
IV. Mechanism of Mg-Proto/Mg-ProtoMe Signaling	294
V. Plastid and Light Signaling Pathways Appear to Interact	297
VI. Conclusions and Perspectives	298
Acknowledgments	296
References	298

Summary

The chloroplast proteome is encoded by genes that reside in both the chloroplast and the nucleus. This separation of genetic material necessitates a system for coordinating the expression of genes that reside in each compartment. Because the overwhelming majority of genes that encode chloroplast proteins reside in the nucleus, the regulation of nuclear genes by developmental and environmental cues plays a dominant role in chloroplast development and function. However, the chloroplast is not indifferent to its own protein composition. In fact, the chloroplast generates signals that have dramatic effects on the expression of nuclear genes that encode particular chloroplast proteins. Currently it is known that plastids produce at least a few distinct signals during chloroplast development that are required for proper expression of particular nuclear genes that encode components of the photosynthetic machinery. In response to certain environmental signals, mature chloroplasts send additional signals that regulate nuclear gene expression. The molecular nature of most of these plastid-to-nucleus signaling pathways is not well established. However, a number of studies have suggested that accumulation of certain chlorophyll precursors within plastids is a signal that regulates nuclear gene expression during chloroplast development and during the diurnal cycle. Future work in this area should provide detailed molecular information on the influence of chlorophyll synthesis and other plastid-localized metabolism on nuclear gene expression and how plants utilize this form of interorganelle communication during their lifecycles.

I. Introduction

The photoautotrophic lifestyle of plants is absolutely dependent on chloroplasts, which are the products of

an endosymbiotic relationship between an ancient eukaryote and a predecessor of modern cyanobacteria. As the endosymbiotic relationship between eukaryotic and cyanobacterial cells gave rise to modern photosynthetic organisms, the overwhelming majority of the ancient cyanobacterial genome was lost or transferred to the nucleus of the eukaryotic host (Herrmann et al., 2003). Currently chloroplast genomes of higher plants are known to encode 60 to 80 proteins, and more than 3500 nuclear genes are predicted to encode chloroplast proteins in *Arabidopsis thaliana* (Martin

*Author for correspondence, email: larkinr@msu.edu

Abbreviations: GUN – Genomes uncoupled; Lhcb – light-harvesting chlorophyll *a/b*-binding protein of photosystem II; Mg-Proto – magnesium protoporphyrin IX; Mg-ProtoMe – magnesium protoporphyrin IX 6-monomethyl ester; Pchl_{id} – protochlorophyllide; Proto – protoporphyrin IX; RBCS – ribulose 1, 5-bisphosphate carboxylase/oxygenase small subunit.

and Hermann, 1998; Arabidopsis Genome Initiative, 2001). As expected, the nucleus plays a dominant role in chloroplast development, but the expression of nuclear genes that encode chloroplast proteins is also dependent on the functional and developmental state of the chloroplast. Chloroplasts send signals to the nucleus that are essential for proper expression of nuclear genes that encode proteins with functions related to photosynthesis, coordinating expression of chloroplast and nuclear genomes, and proper leaf morphogenesis (Rodermel, 2001; Surpin et al., 2002; Gray et al., 2003; Rodermel and Park, 2003; Reinbothe and Reinbothe, this volume). Because chloroplasts perform essential metabolic functions, it is not surprising that the functional state of the chloroplast has a dramatic influence on gene expression and developmental decisions, but little is known about the molecular mechanisms that chloroplasts use to communicate with other cellular compartments.

Mayfield and Taylor (1984) used maize seedlings in which chloroplast development was arrested at an early developmental stage to provide the first evidence that the developmental state of the chloroplast has a powerful effect on the expression of nuclear genes that encode particular chloroplast proteins. Subsequently, a number of studies have indicated that proper expression of nuclear genes that encode proteins with functions related to photosynthesis is dependent on normal chloroplast development in diverse monocotyledonous and dicotyledonous plants (Oelmüller, 1989; Gray et al., 2003; Strand et al., 2003). More recent experiments indicate that there are a number of distinct plastid-to-nucleus signaling pathways. Each of these signaling pathways is likely essential for proper metabolism under particular developmental and environmental conditions (Mochizuki et al., 2001; Rodermel et al., 2001; Mullineaux and Karpinski, 2002; Surpin et al., 2002; Gray et al. 2003; Pfannschmidt, 2003; Rodermel and Park, 2003).

The physiological significance of some chloroplast signaling pathways seems clear. For example, some pathways fine tune the expression of nuclear genes that encode components of the photosynthetic machinery to particular light environments (Pfannschmidt, 2003); other pathways induce the expression of nuclear-encoded antioxidant defense proteins as reactive oxygen species accumulate within the chloroplast (Mullineaux and Karpinski, 2002; Mullineaux et al., this volume). It is likely that plastid signaling pathways linking chloroplast development to other cellular processes are important as seedlings emerge from underneath the soil and/or ground cover and begin the transition from heterotrophic to photoautotrophic growth.

Examples of plastid-to-nucleus signaling pathways that might be important during photomorphogenesis include pathways that link leaf morphogenesis and the expression of nuclear genes that encode proteins active in photosynthesis to chloroplast development (reviewed in Oelmüller, 1989; Rodermel, 2001; Surpin et al., 2002; Gray et al., 2003; Rodermel and Park, 2003; Reinbothe and Reinbothe, this volume). Plastids appear to use different pathways for influencing leaf development and coordinating the expression of nuclear genes encoding proteins that are active in photosynthesis with chloroplast development (Rodermel, 2001). There is now substantial evidence that the cell uses more than one signal to monitor chloroplast development, and one of these signals appears to be the accumulation of particular chlorophyll precursors. In this chapter, the data supporting a role for chlorophyll precursors in intracellular communication are reviewed and current models for chlorophyll precursor signaling as well as future directions of this field are discussed.

II. Chlorophyll Biosynthetic Mutant, Inhibitor, and Feeding Studies

Tetrapyrroles are the intermediates and end products of the chlorophyll, heme, and phytychromobilin biosynthetic pathway (Fig. 1a). Tetrapyrroles are best known for their importance in metabolism, but there is precedence for heme (Fig. 1b) also functioning as a ligand for factors that regulate gene expression in yeast, animal, and bacterial cells (Chen and London, 1995; Sassa and Nagai, 1996; Zhang and Hach, 1999; O'Brian and Thony-Meyer, 2002). Mg-protoporphyrin IX monomethyl ester (Mg-ProtoMe, Fig. 1c), a chlorophyll precursor that bears a striking resemblance to heme, was first suggested to act as a regulator of nuclear gene expression in *Chlamydomonas reinhardtii* (Johanningmeier and Howell, 1984). Using mutations and inhibitors that were previously shown to affect accumulation of the chlorophyll precursor Mg-ProtoMe, these researchers reported an inverse correlation between Mg-ProtoMe accumulation and the levels of the light-harvesting chlorophyll a/b-binding protein of photosystem II (Lhcb) mRNA and ribulose 1, 5-bisphosphate carboxylase/oxygenase small subunit RBCS mRNA (Johanningmeier and Howell, 1984; Johanningmeier, 1988; Jasper et al., 1991). Protoporphyrin IX (Proto) and protochlorophyllide (Pchl) appeared to be less effective regulators of nuclear transcription than Mg-ProtoMe (Johanningmeier and Howell, 1984).