Biochimica et Biophysica Acta xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

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

Review

Translational regulation in chloroplasts for development and homeostasis ☆

Yi Sun, William Zerges *

Biology Department and Center for Structural and Functional Genomics, Concordia University, 7141 Sherbrooke W., Montreal, Quebec H4B 1R6, Canada

ARTICLE INFO

Article history:
Received 15 January 2015
Received in revised form 13 April 2015
Accepted 10 May 2015
Available online xxxx

Keywords:
Translation
Chloroplast
Plastid
Chlamydomonas
Development
Photomorphogenesis

ABSTRACT

Chloroplast genomes encode 100–200 proteins which function in photosynthesis, the organellar genetic system, and other pathways and processes. These proteins are synthesized by a complete translation system within the chloroplast, with bacterial-type ribosomes and translation factors. Here, we review translational regulation in chloroplasts, focusing on changes in translation rates which occur in response to requirements for proteins encoded by the chloroplast genome for development and homeostasis. In addition, we delineate the developmental and physiological contexts and model organisms in which translational regulation in chloroplasts has been studied. This article is part of a Special Issue entitled: Chloroplast biogenesis.

1. Introduction

Chloroplasts contain a few thousand proteins which function in photosynthesis, the biosynthesis of fatty acids, amino acids, hormones, vitamins, nucleotides, secondary metabolites, and intracellular signaling [1]. Most of these proteins are encoded by the nuclear genome, synthesized by cytosolic eukaryotic-type ribosomes, imported into the chloroplast, and targeted to a specific compartment of the chloroplast where they function. Chloroplast genomes encode 100–200 of these proteins, most of which function in photosynthesis and the organellar genetic system. These proteins are synthesized by the chloroplastic translational machinery, which is "bacterial-type" reflecting the evolutionary origin of plastids; a cyanobacterial endosymbiont in the ancient common ancestor of plants and algae. Hence, chloroplastic ribosomes are more like bacterial ribosomes than those of the eukaryotic cytoplasm [2].

A variety of model organisms are used in studies of translational regulation in chloroplasts. The dicot *Arabidopsis thaliana* (hereafter "Arabidopsis") is the most widely used model plant, largely due to its amenability to genetic analyses and the many resources and tools that have been developed by a large research community [3]. Maize is advantageous as a model organism because essential chloroplast processes, like translation, can be dissected with genetic analyses because zygotic lethal mutant seedlings provide sufficient quantities of tissue for analyses of translation defects at the molecular level [4]. Maize and barley seedlings present a developmental gradient of green tissue allowing characterizations of developmental profiles of chloroplast gene expression (Fig. 1A) [5–8]. Lateral sections from seedling leaves have cells in essentially the same stage of differentiation for molecular analyses of chloroplast gene expression [5–8]. Tobacco is the only vascular plant in which the chloroplast genome can be routinely transformed with exogenous DNA, allowing analyses of the effects of site-directed mutagenesis or gene disruptions of translational components, and the use of chimeric reporter gene constructs to monitor translation rates in vivo [9]. The unicellular green alga *Chlamydomonas reinhardtii* (hereafter Chlamydomonas) is used as a model in chloroplast biology because it is amenable to diverse experimental approaches [10]. For example, one can readily monitor translation rates by in vivo radioisotopic pulse labeling, transform the chloroplast genome with exogenous DNA, obtain and study viable mutants deficient in the translation of chloroplast mRNAs encoding photosynthesis proteins [11].

Chloroplast compartments and components are mentioned throughout this review. The chloroplast stroma is the aqueous–proteinaceous compartment that is functionally analogous to the cytoplasm of a cell. The chloroplast envelope transports metabolites, imports chloroplast proteins encoded by the nuclear genome, and synthesizes various chloroplast lipids [12]. Thylakoids are a network of membranous vesicles which carry out the light-dependent reactions of photosynthesis and ATP synthesis

☆ This article is part of a Special Issue entitled: Chloroplast biogenesis.

* Corresponding author. Tel.: +1 514 848 2424x3416.
E-mail address: william.zerges@concordia.ca (W. Zerges).

http://dx.doi.org/10.1016/j.bbabio.2015.05.008
0005-2728/

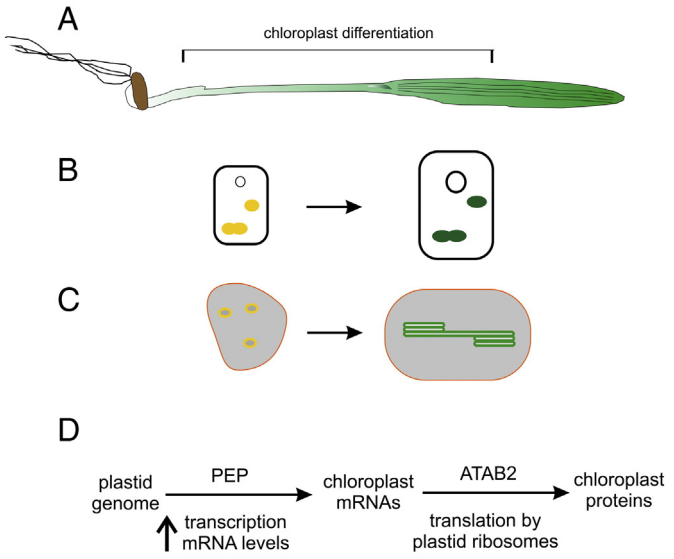

Fig. 1. Translational regulation in chloroplast development from the proplastid. (A) Chloroplast differentiation occurs in cells near the base of the monocot leaf, where meristematic tissue develops into photosynthetic green tissue. (B) In these cells, proplastids (yellow ovals) differentiate to chloroplasts (green ovals). The nucleus is shown as a circle. (C) During proplastid to chloroplast development, prothylakoid vesicles (yellow) differentiate to the thylakoid vesicles (green). (D) This is accompanied by increased chloroplast transcription and mRNA levels. Photosynthesis genes are transcribed primarily by the plastid-encoded RNA polymerase (PEP) [163]. ATAB2 activates the translation of mRNAs encoding subunits of PSI and PSII in Arabidopsis (Section 2) [27].

[13]. Embedded in thylakoid membranes are the multi-subunit complexes of the photosynthetic electron transport system; photosystem I (PSI), photosystem II (PSII), and the cytochrome b6/f complex, as well as the ATP synthase. The CO_2-fixing step in the Calvin cycle is carried out by ribulose bis-phosphate carboxylase–oxygenase (Rubisco); as a multi-subunit "holoenzyme" complex in the chloroplast stroma. Most studies of translational regulation in chloroplasts have focused on genes encoding proteins in photosynthesis.

Translation in chloroplasts has been covered by excellent reviews recently. Some are comprehensive [14–16] while others focus on specific themes; translational regulation by light and redox poise [17,18], translation factors with repeated amino acid sequence motifs (pentatricopeptide repeats (PPR) or octotricopeptide repeats (OPRs)) [17,19], evidence that translation is localized to "biogenesis centers" in chloroplasts and cyanobacteria [20,21], co-translational roles of molecular chaperones [22,23], small regulatory RNAs (sRNAs) [24], and the essentiality of translation in plastids for plant development [25].

Here, we focus on translational regulation in chloroplasts as responds to changing requirements for proteins encoded by the chloroplast genome. We also delineate the different contexts in which this regulation occurs. For example, translational regulation is required for the biogenesis that occurs in chloroplasts as they differentiate from proplastids during the early development of stem and leaf tissues (Section 2). Translational regulation has been studied in chloroplasts differentiating from etioplasts in de-etiolating monocot seedlings (Section 3). Mature chloroplasts regulate translation for the biogenesis that underlies their growth for division in the expanding cells of green tissues and in growing algal populations (Section 4). Translational regulation in chloroplasts also occurs in response to changing environmental conditions, for example, repair the photosynthesis machinery (Section 5). Thus, this review highlights how translational regulation in chloroplasts meets developmental and physiological requirements for proteins encoded by the plastid genome and we discuss the underling regulatory mechanisms.

2. Translational regulation in differentiating chloroplasts in developing green tissues

Proplastids of meristematic cells differentiate to photosynthetic chloroplasts in developing photosynthetic tissues of leaves and stems [26]. This involves the bulk synthesis of most of the polypeptides that function in chloroplasts. Transcriptome profiling of a developmental gradient along the longitudinal axes of the seedling leaves in monocots, e.g., maize and barley (Section 1), revealed drastic increases in chloroplast mRNA levels early in differentiation (Fig. 1) [5–8]. These increases reflect the activation of the plastid-encoded-RNA-polymerase and increased transcription of photosynthesis genes of the plastid genome. However, little is known about translational regulation during proplastid-to-chloroplast differentiation, apart from the role of the RNA-binding protein ATAB2 (Section 2.2).

2.1. ATAB2 is a translational regulator in Arabidopsis

In Arabidopsis, ATAB2 was proposed to regulate translation of chloroplast mRNAs encoding subunits of both photosystems early in proplastid-to-chloroplast differentiation, based on several findings [27]. *ATAB2* expression is induced early in seedling development when proplastid-to-chloroplast differentiation occurs. This induction involves the photoreceptors that initiate development; the cryptochromes. ATAB2 is required for loading the *psaA/B and psbD/C* polycistronic mRNAs on polysomes. Finally, the ATAB2 homologue in Chlamydomonas, Tab2, is required for the translation of the chloroplast *psaB* mRNA [27,28]. Future research of ATAB2 and TAB2 could provide a valuable avenue to elucidate the activation of translation during these earliest stages of chloroplast development, a challenging endeavor considering the tremendous increases in chloroplast mRNA levels and transcription of the chloroplast genome which occur during then (Section 2.1).

3. Translational regulation occurs during etioplast-to-chloroplast differentiation

In angiosperm seedlings that have germinated in the dark, the proplastid differentiates to an etioplast (Fig. 2). Etioplasts lack chlorophyll-binding proteins due to a strict light-requirement for chlorophyll biosynthesis in this phylogeny group [29] and are poised to undergo rapid differentiation to chloroplasts upon illumination and the onset of chlorophyll synthesis, a process called de-etiolation or "greening". Pioneering studies examined translational regulation of plastid gene expression during greening of etiolated seedlings because one can obtain desired amounts of chloroplasts in defined stages of differentiation for molecular and biochemical analyses. Isolated etioplasts were often used because they readily take up exogenous radiolabelled amino acids, allowing determinations of protein synthesis rates (in in organello radioisotope pulse-labeling experiments). Translation rate was inferred from the synthesis rate of a polypeptide normalized to the level of the mRNA encoding it (determined with Northern blot analysis). Translation rates were also determined by the degree of polysome loading of specific mRNAs (determined by sucrose density gradient ultracentrifugation).

3.1. Translational regulation during de-etiolation

De-etiolation activates expression of the chloroplast genes encoding chlorophyll-binding apoproteins of PSII (D1, D2, CP43, CP47) and PSI (PsaA and PsaB) and the LSU of Rubisco [30–33] via translational regulation based on the following. Drastic increases in the rates of radioisotope pulse-labeling of PsbA, PsaA, PsaB, and the LSU of Rubisco were observed in isolated etioplasts as they underwent greening, while levels of the mRNAs encoding these proteins changed less [31–33]. This could be due to repression of the elongation phase of translation in the dark because ribosome occupancy on the initiation

Please cite this article as: Y. Sun, W. Zerges, Translational regulation in chloroplasts for development and homeostasis, Biochim. Biophys. Acta (2015), http://dx.doi.org/10.1016/j.bbabio.2015.05.008