A Genetic Analysis of Chloroplast Division and Expansion in Arabidopsis—Commentary

Pyke KA, Leech RM (1994) A genetic analysis of chloroplast division and expansion in *Arabidopsis thaliana*. Plant Physiol **104:** 201–207

In the early 1990s, nothing was known about the genetic basis of plastid division in photosynthetic eukaryotes. By then, Arabidopsis (*Arabidopsis thaliana*) was becoming well entrenched as a model system for forward genetic analysis, but few mutant screens had centered on problems in the realm of plant cell biology. At that time, Kevin Pyke and Rachel Leech at the University of York undertook a creative and ultimately highly productive microscopic screen for Arabidopsis mutants with defects in the accumulation and replication of chloroplasts, or *arc* mutants. These mutants were initially described in a series of three articles (Pyke and Leech, 1991, 1992; also 1994 article cited above), any of which could be considered a classic. I have chosen to nominate the 1994 article because the mutant characterization was more comprehensive and the images more striking than in the earlier articles.

The screen involved image capture coupled with quantitative analysis of chloroplast size and number as a function of cell size in leaf mesophyll cells (Pyke and Leech, 1991). The 1994 article beautifully illustrated the variations in chloroplast morphology among the *arc* mutants. Some had fewer and larger and some more and smaller chloroplasts than in the wild type; some had uniform chloroplast morphologies, while others had more heterogeneous phenotypes. The screen further revealed that chloroplast division could be radically altered without dramatically impairing plant growth or fertility, indicating that chloroplast division could be dissected genetically. This fact was underscored by the phenotype of *arc6*, which had only one or two giant chloroplasts per cell (Pyke et al., 1994). The quantitative analyses of the *arc* mutants also provided compelling evidence that total chloroplast compartment size as a function of cell size was tightly regulated (Pyke, 1997).

Over time, 12 independent *arc* loci were isolated, nine of which are described in the literature at some level (Marrison et al., 1999; Pyke, 1999). The majority of the mutations, all in nuclear genes, have since been identified. Although some turned out to be alleles of known genes (e.g. *arc11*, Fujiwara et al., 2004; *arc12*, Glynn et al., 2007), map-based cloning yielded several new division proteins. Chief among these were ARC3, which mediates positioning of the cytoskeletal FtsZ ring (Vitha et al., 2001) at the midplastid division site (Shimada et al., 2004; Glynn et al., 2007; Maple et al., 2007), ARC5 (also called DRP5B), a dynamin-related protein that helps constrict the chloroplast from the outside (Gao et al., 2003), and ARC6, an inner envelope protein that both promotes assembly of the FtsZ ring inside the chloroplast and coordinates the FtsZ and ARC5 rings across the envelope membranes (Vitha et al., 2003; Glynn et al., 2008). Through sequence similarity, cloning of ARC6 resulted in the identification of its plastid division paralogue, PARC6 (Glynn et al., 2009). Furthermore, the success of the *arc* screen inspired a later screen for additional *plastid division* mutants, yielding Plastid Division1 (PDV1) and PDV2, outer envelope proteins mediating the recruitment and contractile activity of ARC5 (Miyagishima et al., 2006).

Significantly, analysis of all the currently known chloroplast division proteins in various *arc* mutant backgrounds continues to provide invaluable information on their functional roles in chloroplast division. Thus, the isolation of the *arc* mutants by Pyke and Leech, exemplified by their classic 1994 article in *Plant Physiology*, dramatically influenced our understanding of the composition and function of the chloroplast division machinery in plants and will continue to inform work in this field for many years.

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