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Plant Cytoskeleton: DELLA Connects Gibberellins to Microtubules

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Gibberellins to microtubules: the DELLA connection

A new study reveals that DELLA proteins directly interact with the prefoldin complex, thus regulating tubulin subunit availability in a gibberellin-dependent manner. This finding provides a mechanistic link between the growth promoting plant hormone gibberellin and organization of the cortical microtubule cytoskeleton.

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Plant cells are surrounded by a rigid wall that precludes their movement. Therefore, plant growth and development relies in large part on regulation of the extent and direction of cell expansion. The cortical microtubule cytoskeleton is a key part of the cellular machinery that defines the direction of cell expansion. Cortical microtubules perform this function by organizing cellulose deposition in the wall [1]. The stronger-than-steel cellulose microfibrils act like hoops around a barrel and specify the direction of cell expansion by constraining turgor-driven growth. Plants regulate the pattern of the cortical microtubule array in response to hormonal and environmental signals to modify their growth and adapt to prevailing conditions. Understanding how signals translate to changes in cortical microtubule organization is thus of fundamental importance.

Gibberellins (GAs) are hormones that promote plant growth by stimulating axial cell expansion. The growth stimulatory effect of GA is correlated with increased transverse orientation of the cortical microtubule array [2, 3]. While the signaling pathway for GAs has been extensively studied [4, 5], how these hormones regulate cortical microtubule organization has remained a mystery. A new study by Locascio *et al*. [6] in this issue of *Current Biology* reveals the DELLA proteins as a mechanistic link between GA and cortical microtubule organization.

DELLA-domain containing proteins are negative regulators of GA-dependent processes. These proteins localize to the nucleus and in the absence of GA they restrain plant growth by binding to and consequently inactivating transcription factors and other regulatory proteins [4, 5]. GA relieves this inhibition by triggering degradation of DELLA proteins by the proteasome. Inhibition via interaction is the basic mode of action of DELLA proteins. Locascio *et al*. discovered a new take on this theme when

they found that GAI, one of five *Arabidopsis* DELLA proteins, interacts with prefoldin 3 and 5. Prefoldin 3 and 5 are subunits of the hexameric prefoldin complex, which is an essential part of the chaperone machinery that facilitates assembly of active αβ-tubulin dimers [7]. Interaction of GAI with the prefoldin complex provides the first hint for how GA regulates cortical microtubules.

While DELLA proteins are found in the nucleus, the prefoldin complex normally resides and functions in the cytoplasm. Locascio *et al*. show that the GAI-prefoldin interaction correlates with localization of the prefoldin complex to the nucleus in a GA-dependent manner (Figure 1). Under low GA conditions, prefoldin is found predominantly in the nucleus. In contrast, when GA levels increase, prefoldin is found predominantly in the cytoplasm. This tight correlation indicates that DELLA proteins bind to and sequester the prefoldin complex in the nucleus in the absence of GA. In the presence of GA, DELLAs are degraded and the prefoldin complex shuttles out into the cytoplasm where it can function to produce active tubulin subunits. Importantly, prefoldin subunits that do not directly bind to GAI also localize to the nucleus in a GA-dependent manner, indicating that the GAI-prefoldin interaction does not disrupt the prefoldin complex.

Mutant analysis has shown that loss of prefoldin activity in *Arabidopsis* plants leads to reduced levels of tubulin subunits, disorganized cortical microtubule arrays and reduced plant growth [8, 9]. DELLA-mediated localization of the prefoldin complex to the nucleus represents a rapid and reversible mechanism to regulate prefoldin activity. If sequestering the prefoldin complex to the nucleus prevents its function, then it is expected to reduce the cellular pool of tubulin dimers. Indeed, using an inhibitor of GA

biosynthesis, Locascio *et al*. found that α-tubulin and β-tubulin subunits tend to be monomeric under conditions that lead to prefoldin accumulation in the nucleus. Under these conditions, the cortical microtubule arrays are more disorganized and also less dense, presumably because tubulin levels are limiting.

Regulation of the prefoldin complex is also important for microtubule-dependent processes in animal cells. In particular, prefoldin expression levels correlate to the growth status of animal cells. Furthermore, overexpression of prefoldin complexes has been observed in many types of cancer and is thought to be important to support the high mitotic activity of tumor cells [10, 11]. Whether plants also regulate prefoldin activity via gene expression remains to be determined.

Since GA-induced plant growth can lead to very large increases in cell surface area, this likely creates an increased demand for tubulin in order to form new cortical microtubules needed to organize cellulose deposition across the growing cell surface. Cortical microtubules are thought to self-organize into coaligned arrays through specific interactions between them [12]. An increase in tubulin availability in response to GA might be important to maintain microtubule growth and density required for frequent cortical microtubule interactions and hence array organization. While rapid plant growth in response to GA generally correlates with increased transverse alignment of CMTs, transverse arrays are also found in cells that are not rapidly expanding [13]. In addition, stable transverse alignment of CMTs does not appear to be necessary for maintaining GA-induced plant growth [3]. One possible reason for this discrepancy is that these observations were conducted on cortical microtubules along the outer epidermal surface, which have been found to be highly variable in their organization even during

phases of rapid cell expansion [14, 15]. Cortical microtubule organization along the inner tangential cell surface is reported to be a more faithful indicator of the cell expansion status [14, 15]. The effect of GA on cortical microtubule organization must be accompanied by deposition of new wall material and modification of linkages between wall polymers for sustained growth. Indeed, genome-wide microarray analysis has found that DELLAs regulate expression of genes that encode for proteins involved in cell wall structure and modification [16]. It will be important to determine how these changes in gene expression relate to wall properties and cell growth.

Plant growth rate is modulated by multiple signals including circadian clock, light and hormones. DELLA proteins are emerging as key factors that might serve to integrate multiple inputs to generate a coherent growth output [17-20]. In support of this idea, Locascio *et al*. provide evidence for diurnal oscillation in the nuclear accumulation of DELLA and prefoldin, concomitant with changes in cortical microtubule organization according to the growth status of cells. Together, the available data place DELLA proteins at the nexus of signaling, cortical microtubule organization and cell growth.

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Figure Legend

In the absence of GA, DELLA proteins hold the prefoldin complex (PFD) in the nucleus by direct interaction. Presence of GA leads to destruction of DELLA proteins, which allows the prefoldin complex to go to the cytoplasm leading to increased production of tubulin dimers. Increased tubulin availability correlates with transverse cortical microtubule alignment and cell expansion.