Cell wall polysaccharides are mislocalized to the vacuole in *echidna* mutants.

. Heather E. McFarlane[1,†], Yoichiro Watanabe[1,†], Delphine Gendre[2], Kimberley Carruthers[1], Gabriel Levesque-Tremblay[1], George W. Haughn[1], Rishikesh P. Bhalerao[2] and Lacey Samuels[1,*]

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Abstract

During cell wall biosynthesis, the Golgi apparatus is the platform for cell wall matrix biosynthesis and the site of packaging, of both matrix polysaccharides and proteins, into secretory vesicles with the correct targeting information. The objective of this study was to dissect the post-Golgi trafficking of cell wall polysaccharides using echidna as a vesicle traffic mutant of Arabidopsis thaliana and the pectin-secreting cells of the seed coat as a model system. ECHIDNA encodes a trans-Golgi network (TGN) localized protein, which was previously shown to be required for proper structure and function of the secretory pathway. In echidna mutants, some cell wall matrix polysaccharides accumulate inside cells, rather than being secreted to the apoplast. In this study, live cell imaging of fluorescent protein markers as well as transmission electron microscopy (TEM)/immunoTEM of cryofixed seed coat cells were used to examine the consequences of TGN disorganization in echidna mutants under conditions of high polysaccharide production and secretion. While in wild-type seed coat cells, pectin is secreted to the apical surface, in echidna, polysaccharides accumulate in post-Golgi vesicles, the central lytic vacuole, and ER-derived bodies. In contrast, proteins were partially mistargeted to internal multilamellar membranes in echidna. These results suggest that while secretion of cell wall polysaccharides and proteins at the TGN both require ECHIDNA, different vesicle trafficking components may mediate downstream events in their secretion from the TGN.