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MEMBRANE TRAFFICKING IN PLANTS

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■ **Abstract** Plant membrane trafficking shares many features with other eukaryotic organisms, including the machinery for vesicle formation and fusion. However, the plant endomembrane system lacks an ER-Golgi intermediate compartment, has numerous Golgi stacks and several types of vacuoles, and forms a transient compartment during cell division. ER-Golgi trafficking involves bulk flow and efficient recycling of H/KDEL-bearing proteins. Sorting in the Golgi stacks separates bulk flow to the plasma membrane from receptor-mediated trafficking to the lytic vacuole. Cargo for the protein storage vacuole is delivered from the endoplasmic reticulum (ER), *cis*-Golgi, and *trans*-Golgi. Endocytosis includes recycling of plasma membrane proteins from early endosomes. Late endosomes appear identical with the multivesiculate prevacuolar compartment that lies on the Golgi-vacuole trafficking pathway. In dividing cells, homotypic fusion of Golgi-derived vesicles forms the cell plate, which expands laterally by targeted vesicle fusion at its margin, eventually fusing with the plasma membrane.

CONTENTS

INTRODUCTION

Functional compartmentation of the eukaryotic cell necessitates the exchange of proteins, lipids, and polysaccharides between membrane compartments via transport intermediates. Although many aspects of membrane trafficking are likely to be shared among eukaryotes, the endomembrane system of higher plants displays distinct organizational features that may entail adaptive specializations in membrane trafficking.

Remarkable progress has been achieved in the analysis of plant membrane trafficking over the past several years, using different systems and various approaches. Fluorescent marker proteins have been exploited to visualize endomembrane compartments and the exchange of material between them (Brandizzi et al. 2002a). Such studies were often done in tobacco leaf cells, cell cultures, or *Arabidopsis* leaf cell protoplasts in which genes of interest were transiently expressed. Cell cultures were also used for biochemical studies of cargo sorting. In-planta studies mainly addressed tissue-specific trafficking processes. Finally, the genetic model *Arabidopsis* is increasingly being used for functional studies of trafficking components and pathways in mutant and transgenic plants.

This review summarizes current knowledge about plant membrane trafficking. To set the stage, specific features of the plant endomembrane system are first described. This is followed by a brief overview of the vesicle trafficking machinery in plants, as revealed by the *Arabidopsis* genome sequence. The main part of this review focuses on membrane trafficking pathways and their functional analysis.

Organization of the Plant Endomembrane System

ENDOPLASMIC RETICULUM The endoplasmic reticulum (ER) reaches out from the nuclear envelope to the cortical regions of the cell and may be subdivided into several functionally distinct domains (Staehelin 1997). The ER is the entry point into the endomembrane system for newly synthesized proteins. After removal of the signal peptide by a luminal signal peptidase, soluble proteins are folded by the chaperone BiP (Vitale & Denecke 1999). Improperly folded proteins appear to be recognized by a quality control mechanism and translocated across the ER membrane for degradation in the cytosol (Brandizzi et al. 2003). Both soluble and type I membrane proteins are N-glycosylated with a branched oligosaccharide upon entry into the ER lumen (Vitale & Denecke 1999). *Arabidopsis* mutants defective in N-glycosylation or lacking α-glucosidase I involved in N-glycan processing are embryo lethal (Lukowitz et al. 2001, Gillmor et al. 2002, Boisson et al. 2001). Type II membrane proteins and those with a C-terminal hydrophobic membrane anchor remain largely exposed to the cytosol. ER-resident proteins are retained in the ER or are recycled back from the Golgi apparatus (Vitale & Denecke 1999). Markers for the ER include GFP-HDEL fused to an N-terminal signal peptide and resident proteins

such as the SAR1 GDP/GTP exchange factor SEC12, the luminal chaperone BiP, and the calcium-binding proteins calreticulin and calnexin (Bar-Peled & Raikhel 1997, Irons et al. 2003). In addition to exchanging membrane with Golgi stacks, the ER segregates various membrane compartments for oil or protein sequestrations, some of which merge with protein storage vacuoles (Chrispeels & Herman 2000).

GOLGI APPARATUS The Golgi apparatus is a major sorting station, delivering cargo proteins to multiple destinations. Plant cells have between several and hundreds of Golgi stacks (Staehelin & Moore 1995, Dupree & Sherrier 1998). Each Golgi stack consists of several morphologically distinct cisternae from the *cis* to the *trans* side followed by a *trans*-Golgi network (TGN), which is often less extensive than in animal cells. Golgi stacks are highly mobile and appear to associate with ER strands (Boevink et al. 1998, Nebenführ & Staehelin 2001).

Plant Golgi stacks glycosylate passenger proteins and synthesize noncellulosic polysaccharides for the cell wall (Staehelin & Moore 1995, Dupree & Sherrier 1998). The functional subdivision of Golgi stacks into *cis*-, medial-, and *trans*-cisternae is based on enzyme activities. For example, mannosidase II is thought to modify N-glycans in a medial compartment (Staehelin & Moore 1995, Dupree & Sherrier 1998). Several Golgi markers are green fluorescent protein (GFP) fusions with specific enzymes such as UDP-glucose transporter GONST1, mannosidase I (GmManI), β1,2-*N*-acetylglucosaminyltransferase I (GnTI or NAGTI), β1,2-xylosyltransferase, and mammalian sialyltransferase (ST) (reviewed by Neumann et al. 2003, Saint-Jore-Dupas et al. 2004). There is no clear sequence for retention in Golgi-resident proteins, but the transmembrane domain and the cytosolic tail seem to be involved (Saint-Jore-Dupas et al. 2004).

ENDOSOMES Endosomes are still poorly defined in plants, although early studies described a partially coated reticulum (PCR) as an endosomal compartment (reviewed by Battey et al. 1999). In mammals and yeast, early/sorting endosomes are distinguished from recycling endosomes and multivesiculate late endosomes/prevacuolar compartments by suitable markers (Gruenberg 2001, Raiborg et al. 2003). In plants, some Rab5-type markers appear to label different populations of putative early endosomes, which are numerous and highly mobile (Ueda et al. 2001). Putative early endosomes may correspond to 100–300-nm vesicles that form aggregates and accumulate plasma-membrane proteins in brefeldin A (BFA)-treated *Arabidopsis* root cells (Geldner et al. 2001). Recently, an endocytic multivesiculate compartment was identified as the prevacuolar compartment (PVC) known to be involved in Golgi-vacuole trafficking (Tse et al. 2004). These putative late endosomes are approximately 200–500 nm in size. Thus plant cells may have at least two distinct endosomal compartments: early endosomes involved in sorting and recycling and late endosomes/PVC en route to the lytic vacuole.

DIVERSITY OF VACUOLES Plant vacuoles are diverse in shape, size, content, and function (Marty 1999). Even functionally related vacuoles may vary between tissues and plant species. There are two major functional types of vacuoles: lytic vacuoles and protein-storage vacuoles, that can occur within the same cell (Paris et al. 1996). Both types of vacuoles are also regenerated in evacuolated tobacco leaf cell protoplasts, supporting their distinct identities (Di Sansebastiano et al. 2001). Although functionally distinct, the two types of vacuoles can fuse, giving rise to a large central vacuole.

Lytic vacuoles are equivalent to animal lysosomes or yeast vacuoles, functioning as compartments for degradation and waste storage. Distinct markers for lytic vacuoles often used in trafficking studies are the γ-form of membrane-localized tonoplast intrinsic protein (TIP) (γ-TIP) and the soluble proteins sweet potato sporamin and acidic cysteine protease aleurain (Matsuoka et al. 1995, Paris et al. 1996). Protein storage vacuoles (PSVs) accumulate proteins that are utilized mainly as nutrients during seed germination, such as 2S albumin, and 7S vicilin-type and 11S legumin-type globulins. Storage proteins are degraded by enzymes that have been sequestered in a lytic compartment ("globoid") within the PSV (Jiang et al. 2001). Protein storage vacuoles may also store lectin, chitinase, and glucanase (Paris et al. 1996, Jiang et al. 2000). The PSV membrane contains characteristic proteins such as the α-form of tonoplast intrinsic protein (α-TIP). In vegetative tissues, specialized storage vacuoles (Δ-vacuoles), which are characterized by δ-TIP on their membrane, accumulate vegetative storage protein synthesized in response to developmental or environmental cues (Jauh et al. 1998).

CELL PLATE—A TRANSIENT COMPARTMENT Dividing somatic cells and cellularizing endosperm form a transient compartment called the cell plate, which originates by homotypic fusion of transport vesicles in the center of the division plane and then grows out to the periphery, eventually fusing with the parental plasma membrane (Samuels et al. 1995, Otegui et al. 2001). In cytokinesis following the male meiotic divisions, the plasma membrane grows in by fusion with tubular networks along the division plane. These tubular networks originate by homotypic fusion of membrane vesicles, which have been transported along microtubules to the division plane (Otegui et al. 2004). Thus the initial phase of cytokinesis is essentially the same in the two systems.

VESICLE TRAFFICKING MACHINERY: INFORMATION FROM THE ARABIDOPSIS GENOME

Vesicle Formation

Vesicle formation from a donor membrane involves activation of a small GTPase by its GDP/GTP exchange factor, resulting in the recruitment of coat proteins. Membrane-bound cargo proteins are sorted into the forming vesicle by interaction with coat proteins, whereas soluble cargo proteins are recognized by cargo receptors that in turn interact with coat proteins.

SAR1, ARF GTPases AND THEIR INTERACTORS Recruitment of COPII coat proteins requires SAR1 GTPase and its GDP/GTP exchange factor, the ER-localized type II transmembrane protein SEC12. *Arabidopsis* encodes three SAR1 GTPases and two SEC12 proteins of which one each was isolated by complementation of yeast mutants and shown to associate with the ER (d'Enfert et al. 1992, Bar-Peled & Raikhel 1997, Vernoud et al. 2003).

Small GTPases of the ARF family mediate budding of COPI vesicles from the Golgi complex and budding of adaptor complex/clathrin-coated vesicles in post-Golgi trafficking (Bigay et al. 2003, Zhu et al. 1998, Nie et al. 2003). *Arabidopsis* encodes between 9 and 12 ARF GTPases (Jürgens & Geldner 2002, Vernoud et al. 2003). Six ARFA1 proteins are closely related to yeast and animal class I ARFs, whereas class II and III ARFs are absent. ARF subgroups A to D are plant-specific. ARF GTPases are activated by specific GDP/GTP exchange factors, ARF-GEFs, which transiently associate with membranes (Jackson & Casanova 2000). *Arabidopsis* encodes several large, but no small, ARF-GEFs: three of the GNOM/Gea1/2p/GBF1 (GGG) class and five of the BIG/Sec7p class (Jürgens & Geldner 2002). GNOM acts in endosomal recycling, unlike the yeast and mammalian members of the GGG class. The 15 *Arabidopsis* GTPase-activating ARF-GAPs represent four structurally distinct classes (Vernoud et al. 2003).

COAT PROTEINS COPII-dependent ER-Golgi anterograde traffic involves four coat proteins. SAR1-GAP SEC23 directly interacts with SAR1 and the cargo-binding subunit SEC24 (Barlowe 2003). The two outer coat proteins SEC13 and SEC31 augment GAP activity on SAR1 GTPase. *Arabidopsis* encodes five SEC23, four SEC24, two SEC13 and two SEC31 proteins (Sanderfoot & Raikhel 2003). AtSEC23 associates with the ER (Movafeghi et al. 1999).

COPI vesicles mediating Golgi-ER retrograde traffic have a seven-subunit coat termed coatomer, which consists of four inner (β, γ, δ, and ζ-COP) and three outer (α, β' and ε-COP) proteins (Bonifacino & Lippincott-Schwartz 2003). *Arabidopsis* has single genes for γ-COP and δ-COP and multiple genes for the other COPI subunits (Sanderfoot & Raikhel 2003). γ-COP colocalizes with ARFA1c to the periphery of Golgi stacks (Pimpl et al. 2000, Ritzenthaler et al. 2002).

In post-Golgi traffic, vesicle coats often contain heterotetrameric adaptor-protein (AP) complexes surrounded by clathrin triskelia (Bonifacino & Lippincott-Schwartz 2003). Adaptor protein complexes consist of two large adaptins (γ and $\beta1$ in TGN-localized AP-1, α and $\beta2$ in endocytic AP-2, δ and $\beta3$ in endosomal AP-3, and ε and $\beta4$ in ill-defined AP-4), a medium and a small adaptin ($\mu1$–4 and $\sigma1$–4, respectively). Large subunits of AP complexes interact with clathrin and assembly proteins, μ-adaptins bind cargo via their tail domain (Bonifacino & Lippincott-Schwartz 2003). *Arabidopsis* encodes all four types of AP complexes (Boehm & Bonifacino 2001). However, their composition has not been clarified and thus *Arabidopsis* μ-adaptins were designated μA to μD (Happel et al. 2004). Experimental evidence has been obtained for AP-1 and AP-2 complexes as well as clathrin heavy chain (CHC) and light chain (CLC) proteins, and some assembly proteins (reviewed by Holstein 2002; see below). Other

adaptors such as Golgi-localized GGAs or stonins do not exist in *Arabidopsis* (Boehm & Bonifacino 2001). Scission of clathrin-coated vesicles involves large GTPases of the dynamin family (Danino & Hinshaw 2001). *Arabidopsis* encodes 16 dynamin-related proteins (DRPs) of which subgroups DRP1 and DRP2 play roles in endomembrane dynamics and trafficking (Hong et al. 2003).

In yeast, the retromer complex constitutes the coat of vesicles that traffic the vacuolar receptor Vps10p from the prevacuolar compartment back to the *trans*-Golgi (Pfeffer 2001). Vps35p interacting with the cytosolic tail of Vps10, Vps26p, and Vps29p form the inner subunits, which are coated with Vps17p and Vps5p. *Arabidopsis* encodes eight homologs of four retromer subunits, whereas a Vps17p homolog has not been identified (Sanderfoot & Raikhel 2003).

SORTING SIGNALS AND CARGO RECEPTORS Soluble proteins are sorted by binding to the lumenal domain of *trans*-membrane receptors with short cytoplasmic tails. The ER retention signal H/KDEL and various vacuolar sorting signals of soluble cargo proteins have been well characterized in plants (Hadlington & Denecke 2000). Vacuolar sorting signals include specific sequences present in N-terminal or C-terminal propeptides (NTPP, CTPP) as well as internal sequences or overall structures (Matsuoka & Neuhaus 1999). *Arabidopsis* ERD2 H/KDEL receptors recycle cargo back from the Golgi to the ER (Hadlington & Denecke 2000). *Arabidopsis* encodes at least seven vacuolar sorting receptors (VSRs) of which VSR1/ELP has been functionally characterized (Ahmed et al. 2000, Shimada et al. 2003).

Sorting of membrane proteins also requires receptors. In yeast, Rer1p retrieves type II membrane proteins such as Sec12p by interaction with their transmembrane domain (Sato et al. 2001). *Arabidopsis* encodes three RER1 proteins with possibly homologous functions (Sato et al. 1999). Tyrosine motifs to be recognized by μ-adaptins are present on plant vacuolar sorting receptors (Saint-Jore-Dupas et al. 2004). Acidic di-leucine motifs mediate endosomal/lysosomal sorting in mammals (Bonifacino & Traub 2003). The role of this motif in plants was analyzed in targeting of membrane-localized endo-1,4-β-glucanase KOR to the cell plate (Zuo et al. 2000).

Vesicle Fusion

Vesicle fusion with the target membrane occurs after the vesicle has shed its coat. Initially, a Rab GTPase on the vesicle membrane interacts with a tethering protein complex on the target membrane. Then SNARE proteins residing on the opposite membranes form a *trans*-complex, which results in membrane fusion.

Rab GTPases AND EFFECTORS Rab GTPases and their effector proteins tether vesicles to target membranes (Whyte & Munro 2002). *Arabidopsis* encodes 57 Rab GTPases grouped into eight subfamilies, A–H, which were assigned tentative roles by sequence homology to yeast and mammalian counterparts (Rutherford & Moore 2002, Vernoud et al. 2003). However, only a few plant Rab GTPases have

been functionally characterized (see below). Several tethering complexes studied in yeast and animals have putative homologs in *Arabidopsis*, including the exocyst at the plasma membrane, TRAPP at the *cis*-Golgi, VFT/GARP at the *trans*-Golgi, C-VPS at the vacuolar membrane, and several components of the Vps34/COG complex involved in Golgi retrograde transport (Jürgens & Geldner 2002, Elias et al. 2003).

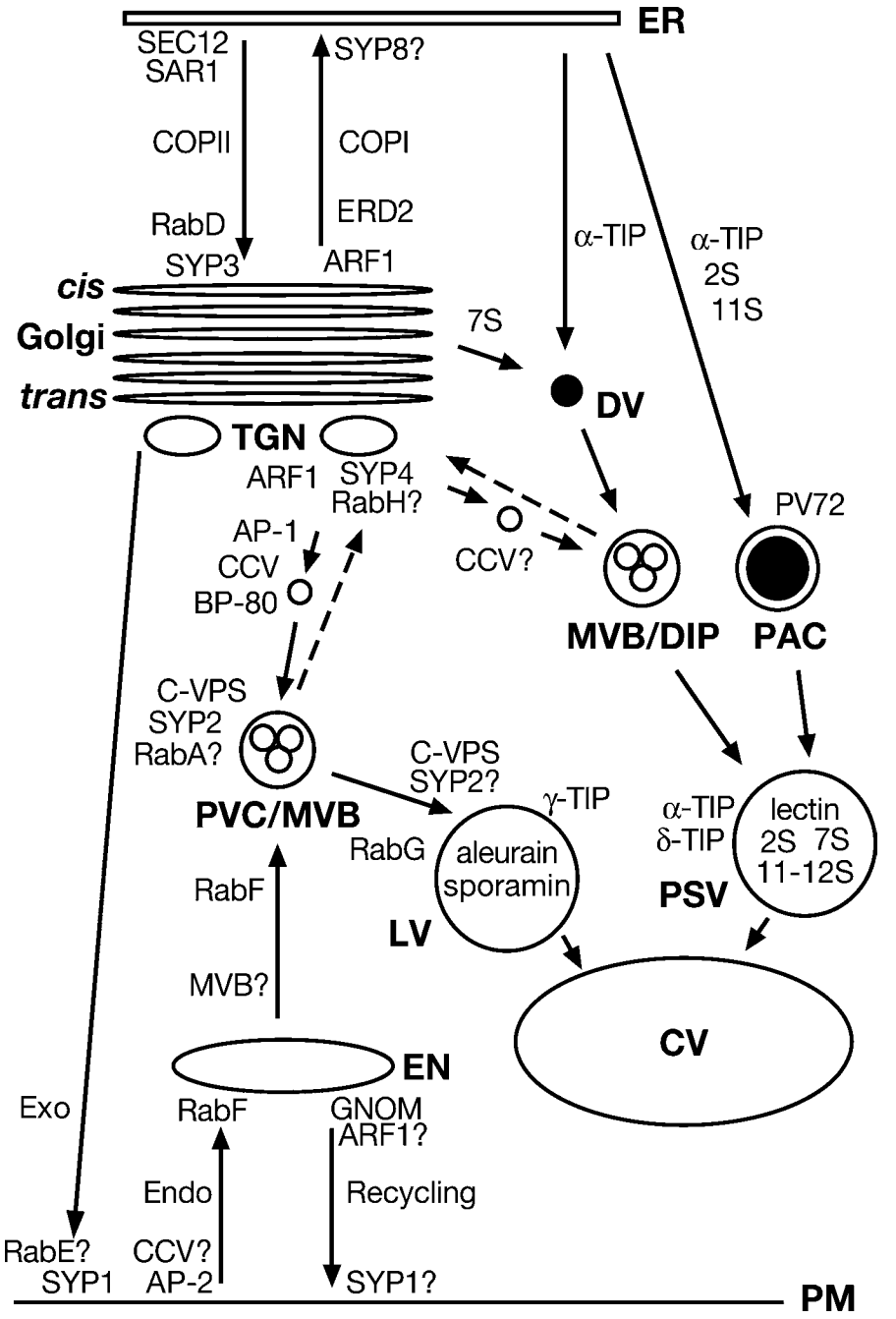
SNAREs AND ASSOCIATED PROTEINS SNARE proteins initiate membrane fusion by forming a *trans*-complex via their distinct R- or Q-SNARE motifs (Jahn et al. 2003). As a rule, an R-SNARE on the vesicle pairs up with two or three Q-SNAREs on the target membrane: a syntaxin (Qa-SNARE) plus either a SNAP25-like protein with two SNARE motifs (Qb,c-SNARE) or two SNARE light chains (Qb-SNARE and Qc-SNARE). Each component of a SNARE complex is a member of a protein family, and different SNARE complexes are involved in different trafficking pathways. *Arabidopsis* encodes at least 54 SNAREs: 18 syntaxins/Qa-SNAREs, 11 Qb-SNAREs, 8 Qc-SNAREs, 3 SNAP25-like/Qb,c-SNAREs, and 14 VAMPs/R-SNAREs. Previously 24 syntaxins of plant (SYPs) were grouped into eight subfamilies by sequence similarity to yeast and mammalian syntaxins (Sanderfoot et al. 2000). All *Arabidopsis* R-SNAREs are "longins" with a characteristic N-terminal domain (Filippini et al. 2001) and, with the exception of SEC22 and YKT6, are related to mammalian endosomal VAMP7 (Sanderfoot et al. 2000). Sec1/Munc18 (SM) family proteins are cytosolic interactors of SNARE proteins and contribute to the specificity of membrane fusion (Jahn et al. 2003). *Arabidopsis* encodes 6 SM proteins: SLY1, VPS33, VPS45, and 3 SEC1-like proteins (Sanderfoot et al. 2000). Following membrane fusion, α-SNAP and the AAA-ATPase *N*-ethylmaleimide-sensitive factor (NSF) assist in the disassembly of *cis*-SNARE complexes (Jahn et al. 2003). Experimental evidence for NSF and α-SNAP action in plants has been obtained for prevacuolar vesicle fusion and cell plate formation (Bassham & Raikhel 1999, Rancour et al. 2002).

TRAFFICKING PATHWAYS

A secretory default pathway leads from the ER via Golgi stacks to the plasma membrane (Figure 1). Storage proteins can bypass the Golgi altogether or exit from the *cis*-Golgi en route to storage vacuoles via intermediate compartments. Recycling and sorting takes place in Golgi stacks, at the PVC en route to the lytic vacuole, and at some endosomal compartment. The endocytic pathway converges on the pathway from the *trans*-Golgi to the lytic vacuole at the PVC.

ER-Golgi Trafficking

There is no evidence for an ER-Golgi intermediate compartment in plants (Neumann et al. 2003). Golgi stacks move over ER strands in an acto-myosin dependent manner (Boevink et al. 1998, Brandizzi et al. 2002b, Saint-Jore et al. 2002, Nebenführ & Staehelin 2001). However, this movement is not required for


ER
SEC12
SAR1
SYP8?
COPII
COPI
RabD
SYP3
ERD2
ARF1
α-TIP
α-TIP
2S
11S
cis
Golgi
trans
7S
DV
TGN
ARF1
SYP4
RabH?
PV72
AP-1
CCV
BP-80
CCV?
MVB/DIP
PAC
C-VPS
SYP2
RabA?
C-VPS
SYP2?
γ-TIP
α-TIP
δ-TIP
lectin
2S 7S
11-12S
PVC/MVB
RabG
aleurain
sporamin
PSV
RabF
LV
MVB?
CV
EN
Exo
RabF
GNOM
ARF1?
Endo
Recycling
RabE?
SYP1
CCV?
AP-2
SYP1?
PM

ER-Golgi trafficking. Rather, ER and Golgi stacks appear to be one dynamic system for the exchange of molecules (Brandizzi et al. 2002b). Disruption of Golgi stacks by BFA treatment resulted in the redistribution of *cis*-Golgi, but not TGN, markers into the ER. On removal of BFA, Golgi stacks were reformed in the presence of protein synthesis inhibitors and cytoskeleton-depolymerizing drugs (Saint-Jore et al. 2002). After photobleaching of individual Golgi stacks, cycling of Golgi-localized GFP markers led to rapid recovery of fluorescence (Brandizzi et al. 2002b). Brefeldin A inhibited the recovery of fluorescence, suggesting that the formation of new Golgi cisternae requires the fusion of Golgi-derived COPI membranes with ER-derived COPII membranes (reviewed by Ward & Brandizzi 2004).

ANTEROGRADE TRAFFIC SAR1 GTPase and its GDP/GTP exchange factor SEC12 mediate COPII-dependent cargo export from the ER (Murshid & Presley 2004). In plants, COPII vesicles have not been observed by electron microscopy. However, GTP-trapped AtSAR1[H74L] caused ER-accumulation of Golgi markers, the vacuolar marker sporamin and the storage protein phaseolin, in transient expression assays (Takeuchi et al. 2000, Sohn et al. 2003, Park et al. 2004). AtSAR1[H74L] also inhibited α-amylase secretion, and the same effect was caused by overexpression of AtSEC12 (Phillipson et al. 2001). Soluble cargo appears to exit the ER by bulk flow in a COPII-dependent manner, with ER proteins being efficiently retrieved from the Golgi complex (Crofts et al. 1999, Phillipson et al. 2001). It is not known whether bulk flow also applies to membrane proteins. In yeast, COPII coat protein Sec24p was shown to select specific types of membrane protein cargo (Barlowe 2003).

Transient expression of dominant-negative AtRAB1b[N121I] resulted in the ER accumulation of secretory GFP (secGFP), which was counteracted by wild-type AtRAB1b but not by AtRAB8c (Batoko et al. 2000). In BFA-treated cells, AtRAB1b[N121I] significantly slowed down the recovery of Golgi-localized

Figure 1 Simplified diagram of plant endomembrane system and trafficking pathways. Secretory cargo is transported from the endoplasmic reticulum (ER) via the Golgi/TGN to the plasma membrane (PM). Cargo destined for the lytic vacuole (LV) is sorted in a BP-80 dependent fashion into clathrin-coated vesicles (CCV) at the TGN and transported to the prevacuolar compartment (PVC/MVB). Cargo destined for the protein-storage vacuole (PSV) is trafficked from the ER to intermediate compartments (DV, PAC, MVB/DIP), from the *cis*-Golgi via dense vesicles (DV), and from the *trans*-Golgi via clathrin-coated vesicles (CCV). PSV and LV may fuse to give a large central vacuole (CV). The endocytic pathway involves an early/sorting endosome (EN) for recycling of plasma-membrane proteins and a multivesiculate late endosome corresponding to the PVC. Vacuolar markers, cargo receptors (ERD2, BP-80, PV72), coat proteins (COPI, COPII, AP-1, AP-2), SAR1, ARF, and Rab GTPases, and syntaxins (SYP) are indicated. DIP, DIP organelle; Endo, endocytosis; Exo, exocytosis; MVB, multivesicular body; PAC, precursor-accumulating compartment. For details, see text.

ST-GFP fluorescence (Saint-Jore et al. 2002). Taken together, anterograde traffic involves SAR1/COPII-dependent export from the ER and RAB1-mediated fusion of cargo carriers at the *cis*-Golgi. The nature of the carriers remains to be determined.

RETROGRADE TRAFFIC Both ER-resident calreticulin and secretory α-amylase fused to HDEL were detected in COPI vesicles isolated from tobacco plants (Pimpl et al. 2000). COPI vesicles, ARF1 GTPase, and COPI coat proteins accumulated at the *cis*-Golgi, suggesting that COPI vesicles mediate retrograde traffic (Pimpl et al. 2000, Ritzenthaler et al. 2002). In BFA-treated BY-2 cells, γ-COP (SEC21) and ARF1 were released into the cytosol (Pimpl et al. 2000, Ritzenthaler et al. 2002). Dominant-negative ARF1 affected ER-Golgi traffic in a manner similar to the action of BFA (Takeuchi et al. 2002, Lee et al. 2002). Thus ARF1 mediates formation of COPI vesicles as carriers in Golgi-ER retrograde trafficking.

Soluble proteins with a C-terminal H/KDEL motif are captured by H/KDEL receptors such as ERD2 for sorting into COPI vesicles. AtERD2-GFP accumulated at Golgi stacks of tobacco cells, as expected for Golgi-to-ER recycling (Boevink et al. 1998). *Arabidopsis* ERD2 proteins have a C-terminal KKXX or KXKXX motif, which mediates recycling of membrane proteins to the ER in yeast and mammalian cells (Jackson et al. 1993). Functionality of the KKXX motif in plants was demonstrated by mutation to NNXX (Benghezal et al. 2000).

Post-Golgi Trafficking

The plant Golgi apparatus is a major sorting station from which trafficking pathways diverge to different types of vacuoles and to the plasma membrane. Trafficking to the protein storage vacuole may also involve an ER-derived route that bypasses the glycan-modifying compartment of the Golgi complex.

PATHWAYS CONVERGING ON THE PROTEIN STORAGE VACUOLE During pea cotyledon development, PSVs are formed de novo from tubular-cisternal precursors, which surround the shrinking vegetative vacuoles. These precursors accumulate vicilin and legumin storage proteins and contain the PSV membrane marker α-TIP (Hoh et al. 1995). These markers were also detected in $\sim$150-nm electron-dense vesicles (DV) without a clathrin coat; DVs formed in the *cis*-Golgi, were carried through the stack and were released from the TGN (Hohl et al. 1996, Hillmer et al. 2001). Isolated dense vesicles lacked γ-COP and the vacuolar sorting receptor BP-80 involved in trafficking to the lytic vacuole (Hinz et al. 1999). Evidence for the two independent trafficking routes to the PSV was obtained in several plant species. In tobacco seeds, TIP-related dark intrinsic protein (DIP) accumulated in 200-nm compound vesicles and in 1–2 μm large-PSV precursors (DIP organelles). These DIP organelles contained α-TIP and a type I membrane protein, RMR, with a Golgi-glycosylated lumenal domain (Jiang et al. 2000).

In pumpkin cotyledon cells, membrane-bounded aggregrates of 2S albumin and 11S globulin precursors segregated from the ER to form the electron-dense core of precursor-accumulating compartment (PAC) vesicles 300–400 nm in diameter (Hara-Nishimura et al. 1998). However, PAC vesicles also had an electron-translucent outer layer containing glycoproteins with Golgi-modified glycans. In leaf cell protoplasts of three plant species, α-TIP bypassed the Golgi complex, as indicated by the lack of Golgi-modified glycans and by the failure of dominant-negative Rab1 to inhibit α-TIP trafficking (Park et al. 2004). In contrast, the storage protein phaseolin from common bean contained Golgi-modified glycans. Furthermore, its traffic was sensitive to BFA and was inhibited by dominant-negative variants of SAR1 and Rab1, indicating that phaseolin is transported via the Golgi complex.

A type I membrane protein, PV72, isolated from PAC vesicles of pumpkin seed was proposed to function as a sorting receptor for storage proteins (Shimada et al. 2002, Watanabe et al. 2002). PV72 is a member of the family of vacuolar sorting receptors (VSRs) and is most closely related to VSR1/AtELP among seven *Arabidopsis* VSRs analyzed (Shimada et al. 2003). Knockout mutants lacking VSR1/AtELP, but not any other VSR, secreted precursors of 12S globulin and 2S albumin storage proteins into the extracellular space, whereas processing of the aleurain precursor destined for the lytic vacuole was not impaired in *vsr1* mutant embryos (Shimada et al. 2003). These results suggest that VSR1/AtELP may act as a VSR for storage proteins in *Arabidopsis* embryos, which is in conflict with evidence for a role of AtELP in trafficking to the lytic vacuole in vegetative cells (see below). Moreover, the lumenal domain of PV72 fused to HDEL trapped aleurain in the ER of transgenic *Arabidopsis* leaves, suggesting that PV72 recognizes the same vacuolar targeting signal as the lytic cargo receptor (Watanabe et al. 2004). To clarify the roles of VSR family members, trafficking to both protein-storage and lytic vacuoles should be analyzed in each of the *Arabidopsis vsr* mutants.

PATHWAY TO THE LYTIC VACUOLE Soluble protein cargo destined for the lytic vacuole includes aleurain and sporamin, which were mainly used in vacuolar trafficking studies. The N-terminal sorting determinant (NTPP) of their precursor forms was recognized by the lumenal portion of VSRs such as pea BP-80 and AtELP, in contrast to the C-terminal targeting determinant (CTPP) of barley lectin (Kirsch et al. 1996, Cao et al. 2000, Ahmed et al. 2000). AtELP also colocalized with sporamin, but not with barley lectin (Ahmed et al. 2000).

All members of the VSR family share a tyrosine-based sorting motif, YMPL (Saint-Jore-Dumas et al. 2004). YMPL of AtELP bound in vitro more efficiently to mammalian μ1-adaptin of TGN-localized adaptor complex AP-1 than to μ2-adaptin of endocytic AP-2 (Sanderfoot et al. 1998). Recently, YMPL of BP-80 was shown to bind in vitro to μA-adaptin from *Arabidopsis*, which localized to the *trans*-face of Golgi stacks and thus appears to be a subunit of the *Arabidopsis* AP-1 complex (Happel et al. 2004). AP-1 specific γ-adaptin interacted in vitro with

the dynamin-related protein DRP2A (ADL6), which localized to clathrin-coated vesicles budding from the *trans*-Golgi (Lam et al. 2002). Dominant-negative ADL6 inhibited trafficking of sporamin to the vacuole but not trafficking of H^+-ATPase to the plasma membrane (Jin et al. 2001). Taken together, these results suggest that AP-1/clathrin-coated vesicles carry cargo destined for the lytic vacuole. Dominant-negative ARF1 inhibited vacuolar trafficking mediated by the BP-80 cargo receptor, implicating ARF1 in the formation of AP-1/clathrin-coated vesicles (Pimpl et al. 2003).

BP-80 and AtELP were localized to both Golgi stacks and the PVC (Ahmed et al. 2000, Paris et al. 1997). Vesicle fusion may involve syntaxin SYP21/PEP12, which localized at the PVC and was shown to reside in a 20S complex (da Silva Conceicao et al. 1997, Bassham et al. 1999). PEP12/SYP21 and syntaxin VAM3/SYP22 formed two related SNARE complexes with Qc-SNARE SYP51 and Qb-SNARE VTI11 (VTI1a) at the PVC (Sanderfoot et al. 2001a). Although PEP12 and VAM3 are closely related, knockouts for each gene were gametophytic lethal, indicating functional diversity (Sanderfoot et al. 2001b). Mutations affecting VTI11 (*zig*) or VAM3 (*sgr3*) caused defects in shoot gravitropism, indicating that the VAM3/VTI11 complex mediates vacuolar trafficking in gravity-sensing shoot endodermis cells (Yano et al. 2003). *zig* mutant plants were viable because VTI12 substituted for VTI11 in SNARE complex formation at the PVC, whereas the *zig vti12* double mutant was embryo lethal (Surpin et al. 2003).

In yeast, vacuolar trafficking requires the class C-Vps complex (Peterson & Emr 2001). The *Arabidopsis* homolog of Vps16, VCL1, is required for vacuole biogenesis. *vcl1* mutant embryos were lethal, forming autophagosomes and secreting aleurain into the extracellular space (Rojo et al. 2001). VCL1 formed a complex with VPS11 and VPS33, which are also homologs of yeast C-Vps proteins, and all three proteins localized to the PVC and the vacuolar membrane (Rojo et al. 2003). Co-immunoprecipitation with anti-VCL1 antiserum suggested that SYP21 and SYP22 syntaxins interact with the putative C-VPS complex. Yeast C-Vps interacts with RabG class Ypt7 (Wurmser et al. 2000). A GFP fusion of a RabG class protein, OsRab7 from rice, was recently localized to the vacuolar membrane in *Arabidopsis* protoplasts (Nahm et al. 2003). Two RabA proteins from pea, RabA3/Pra2 and RabA4/Pra3, may also be involved in Golgi-PVC/endosomal trafficking but their precise roles need to be determined (Inaba et al. 2002).

RETROGRADE TRAFFIC FROM THE PREVACUOLAR COMPARTMENT Vacuolar sorting receptors need to be recycled to the *trans*-Golgi after releasing their cargo into the PVC. Fusion of PVC-derived vesicles may involve SNARE complexes that were localized to the TGN in *Arabidopsis* (Sanderfoot et al. 2001a). Two of these complexes contained Qb-SNARE VTI12, Qc-SNARE OSM1/SYP61, and the related syntaxins, either SYP41 (TLG2a) or SYP42 (TLG2b). The SM protein VPS45 localized to the TGN and interacted with both TLG2a and TLG2b, but not with

the PVC-localized syntaxins PEP12 and VAM3 (Bassham et al. 2000). Although TLG2a and TLG2b are closely related, knockouts for each gene were gametophytic lethal, indicating functional diversity (Sanderfoot et al. 2001b). A mutation in *SYP61* (*osm1*) caused hypersensitivity to osmotic stress (Zhu et al. 2002). Elimination of VTI12 accelerated senescence under poor growth conditions, suggesting a possible role of VTI12 in autophagy (Surpin et al. 2003). VTI11 substituted for VTI12 in TGN-localized SNARE complexes of *vti12* mutants, the double mutant being embryo lethal.

Trans-GOLGI TO PLASMA MEMBRANE—DEFAULT PATHWAY The transport route from the *trans*-Golgi to the plasma membrane appears to be the default pathway for soluble cargo, as demonstrated for a secreted form of GFP (secGFP; Batoko et al. 2000). The *Arabidopsis* peptide ligand CLV3, which normally spreads into adjacent cells, was diverted to the vacuole when fused to a vacuolar sorting signal (Rojo et al. 2002, Lenhard & Laux 2003). How membrane proteins are targeted to the plasma membrane is not known.

Transport vesicles destined for the plasma membrane have not been unambiguously identified in plants. Nonetheless, several syntaxins and SNAP25 homologs were localized at the plasma membrane, including *Arabidopsis* SYP121/PEN1 and its barley ortholog ROR2 (both of which are involved in nonhost pathogen resistance); their putative tobacco homolog SYR1; SYP122, possibly involved in response to a bacterial elicitor; and *Arabidopsis* SNAP33 and barley SNAP34 (Collins et al. 2003, Leyman et al. 2000, Nühse et al. 2003, Heese et al. 2001). Overexpression of the cytosolic fragment of SYR1 prevented secretion of secGFP, which then accumulated in the ER and Golgi stacks, and this inhibitory effect was abolished by simultaneous overexpression of SYR1 (Geelen et al. 2002).

Several *Arabidopsis* plasma-membrane proteins were localized in specific subdomains. The putative auxin-efflux carrier PIN1 accumulated at the basal surface of vascular cells, the auxin-influx carrier AUX1 at the apical surface, GPI-anchored COBRA protein at the lateral surfaces in inner root cells, and PIN2 protein at the apical surface of root epidermis cells (reviewed by Jürgens & Geldner 2002). Targeted trafficking to plasma-membrane subdomains requires sorting from the general bulk flow, which may occur in Golgi stacks. Alternatively, targeted trafficking may involve recycling from endosomes (see below).

Endocytosis

In yeast and animals, receptor-ligand complexes are internalized for signaling, for dissociation of the complex and receptor recycling, or for eventual degradation in the lysosome/vacuole (Clague & Urbe 2001). Information on receptor-mediated endocytosis is still scant in plants, although several receptor kinases and their peptide ligands have been identified (Matsubayashi 2003). SERK1 receptor kinase was internalized on dephosphorylation by a kinase-associated protein phosphatase

(Shah et al. 2002). A sorting nexin, SNX1 from *Brassica*, interacted in vitro with the kinase domains of SRK and CLV1, suggesting trafficking of these receptors through the endosomal system (Vanoosthuyse et al. 2003).

Early studies of plant endocytosis made use of electron-dense markers such as cationized ferritin; its uptake into protoplasts and intracellular trafficking to the vacuole were analyzed by electron microscopy (reviewed by Battey et al. 1999). More recent studies employed the endocytic tracers FM1-43 and FM4-64 (Emans et al. 2002, Tse et al. 2004). *Arabidopsis* αC-adaptin, a subunit of a putative endocytic AP-2 complex, interacted with clathrin and the assembly factor homolog AtAP180, and AtAP180 was shown to mediate clathrin assembly into regular cages (Barth & Holstein 2004). Thus there is accumulating evidence for endocytosis and its machinery in plants (reviewed by Holstein 2002).

Trafficking from Endosomes

Early or sorting endosomes are not well characterized in plants. Early studies described a partially coated reticulum (PCR) with clathrin-coated buds (reviewed by Battey et al. 1999). In animals, Rab5 GTPase has been established as a convenient marker for early endosomes, and its *Arabidopsis* homologs RabF1 (Ara6) and RabF2b (Ara7) were shown to colocalize with FM4-64 in endomembrane compartments after brief uptake periods (Ueda et al. 2001). Treatment with BFA led to BFA compartments as vesicle aggregates, which accumulated the *Arabidopsis* BFA-sensitive ARF-GEF GNOM, PIN1, and other plasma-membrane proteins, sterols, and cell-wall pectins (Steinmann et al. 1999, Geldner et al. 2001, Baluska et al. 2002, Grebe et al. 2003). GNOM also colocalized with FM4-64, and Ara7-positive compartments were structurally altered in *gnom* mutant protoplasts (Geldner et al. 2003). Internalized filipin-labeled sterols were colocalized with Ara6 (Grebe et al. 2003). The BFA compartments were clearly distinct from Golgi stacks, as indicated by ultrastructural analysis and double labeling with several Golgi markers (Geldner et al. 2001, 2003; Baluska et al. 2002; Grebe et al. 2003).

RECYCLING TO THE PLASMA MEMBRANE Recycling of plasma-membrane proteins is thought to involve sorting in the early endosome (Woodman 2000). In plants, recycling of plasma-membrane proteins was demonstrated by washing out BFA in the presence of cycloheximide (Geldner et al. 2001). Similarly, internalized sterol was recycled to the plasma membrane in an actin-dependent manner (Grebe et al. 2003). In plants expressing BFA-insensitive GNOM, PIN1 did not accumulate in BFA compartments, whereas PIN2 and H^+-ATPase were still partially sensitive to BFA (Geldner et al. 2003). H^+-ATPase-GFP accumulated in Golgi marker-negative BFA compartments on overexpression of dominant-negative AtARFA1c (Lee et al. 2002). BFA compartments of maize root cells were labeled by anti-AtARFA1c antiserum (Baluska et al. 2002). As *Arabidopsis* ARF1 proteins are

almost indistinguishable, the identity of the endosomal ARF1 protein(s) remains uncertain. Taken together, these results suggest multiple recycling pathways.

TRAFFICKING TO THE LYTIC VACUOLE In animal and yeast cells, proteins destined for degradation in the lysosome/vacuole are sorted into internal vesicles of the early endosome (Raiborg et al. 2003). The multivesicular body (MVB) thus formed detaches from the endosome and fuses with the late multivesicular endosome/prevacuolar compartment. In plants, endocytic trafficking from the early compartment described above appears to involve a multivesiculate late compartment, which was labeled with both FM4-64 and the vacuolar sorting receptor BP-80 (Tse et al. 2004). In addition, BP-80 colocalized with the prevacuolar syntaxin PEP12 (SYP21), indicating that the late compartment corresponds to the PVC that lies on the trafficking route from the *trans*-Golgi to the lytic vacuole (see above). Wortmannin, an inhibitor of phosphatidylinositol 3-kinase (PI3K), blocked FM1-43 traffic to the vacuole (Emans et al. 2002). Wortmannin also affected the morphology of the multivesiculate PVC (Tse et al. 2004). Transiently expressed phosphatidylinositol 3-phosphate (PI3P)-binding FYVE domain from human EEA1 fused to GFP labeled the PVC, as indicated by colocalization with BP-80, and overexpression of the FYVE domain inhibited vacuolar targeting of sporamin (Kim et al. 2001). These results suggest that PI3P plays a role in trafficking at the PVC.

Arabidopsis has three RabF GTPases related to endosomal Rab5 in mammals. RabF1 (Ara6) and RabF2b (Ara7) were localized to different subpopulations of endosomes. Both GTP-locked Ara6 and Ara7 caused fusion of endosomes to form larger structures, whereas GDP-locked Ara6[N147I] resided on the plasma membrane (Ueda et al. 2001). Dominant-negative Ara6 did not interfere with vacuolar trafficking of sporamin, in contrast to dominant-negative RabF2a (Rha1) and Ara7 (Sohn et al. 2003). Thus Ara6 seems to act before the endocytic pathway converges on the vacuolar trafficking pathway. In contrast, a putative Ara6 ortholog from *Mesembryanthemum*, called m-Rab_{mc}, was predominantly localized to the PVC, and dominant-negative m-Rab_{mc} blocked vacuolar targeting of aleurain presumably at the PVC (Bolte et al. 2004). In addition, m-Rab_{mc}[N147I] was cytosolic, in contrast to the plasma-membrane localization of Ara6[N147I] (see above). Thus the putative orthologs Ara6 and m-Rab_{mc} appear to have different roles in endocytic trafficking. The third Rab5-related *Arabidopsis* RabF GTPase, Rha1, colocalized with the PVC markers VSR1/ELP and PEP12 (SYP21), and dominant-negative Rha1 blocked delivery of sporamin and aleurain to the lytic vacuole at the PVC (Sohn et al. 2003).

Trafficking During Cytokinesis

Cytokinesis of somatic cells progresses from the center to the periphery and is assisted by a dynamic cytoskeletal array, the phragmoplast, which delivers membrane

vesicles to the division plane, probably along microtubules (Staehelin & Hepler 1996, Otegui et al. 2001). Initially, homotypic vesicle fusion generates a network of tubular membranes, which is then transformed into a disk-shaped continuous membrane compartment, the cell plate, that secrets callose into its lumen (Samuels et al. 1995, Otegui et al. 2001). The tubular membranes are locally constricted by dynamin-related protein DRP1a, which may also localize callose synthase (Otegui et al. 2001; Hong et al. 2001a,b). An *Arabidopsis* double mutant lacking both DRP1a (ADL1A) and related DRP1e displayed defects in cell plate formation (Kang et al. 2001, 2003).

Lateral translocation of the phragmoplast targets later-arriving vesicles to the margin of the cell plate, which thus grows toward the cell periphery, eventually fusing with the plasma membrane. In vacuolate cells, the cell plate appears to fuse with the plasma membrane locally before spreading along the cell margin to the opposite side (Cutler & Ehrhardt 2002). Although unique, plant cytokinesis bears some resemblance to the final stage of animal cytokinesis during which the midbody gap is closed by vesicle delivery and SNARE-mediated vesicle fusion (Low et al. 2003).

Trafficking to the plane of division appears to be the default pathway in dividing cells, as suggested by the accumulation of plasma-membrane proteins such as PIN1 and KOR, and secretory proteins such as endoxyloglucan transferase (Steinmann et al. 1999, Zuo et al. 2000, Yokoyama & Nishitani 2001). This trafficking probably results from bulk flow and targeted delivery of Golgi-derived vesicles along phragmoplast microtubules.

CELL PLATE FORMATION BY VESICLE FUSION Unfused transport vesicles 60–80 nm in diameter accumulated in *Arabidopsis* cytokinesis mutants lacking the cytokinesis-specific syntaxin KNOLLE or its interactor, the SM protein KEULE (Lauber et al. 1997, Waizenegger et al. 2000, Assaad et al. 2001). A KNOLLE-interacting SNAP25 homolog, SNAP33, colocalized with KNOLLE at the cell plate but also accumulated at the plasma membrane in nondividing cells (Heese et al. 2001). Thus the cytokinetic SNARE complex contains a specific syntaxin and a promiscuous SNAP25 homolog. A plant-specific Qb-SNARE, NPSN11, was also localized to the cell plate and appeared to coimmunoprecipitate with KNOLLE (Zheng et al. 2002). An *npsn11* knockout mutant was viable, possibly because of functional redundancy. If NPSN11 formed a complex with KNOLLE and SNAP33, this SNARE complex would be rather unusual, consisting of Q-SNAREs only. Alternatively, KNOLLE may form several SNARE complexes with different partners. Another syntaxin localized to the division plane was the Sed5 homolog SYP31, which labeled Golgi membranes in nondividing cells (Rancour et al. 2002). SYP31 and KNOLLE interacted in vitro with different AAA-ATPases, CDC48 and NSF, respectively. These results were interpreted as evidence for two distinct fusion pathways during cytokinesis (Rancour et al. 2002).

Syntaxin specificity in cytokinesis was studied by expressing several syntaxins under the control of *KNOLLE cis*-regulatory gene sequences (Müller et al. 2003).

Both prevacuolar PEP12 (SYP21) and plasma membrane-localized PEN1/SYR1 (SYP121) failed to rescue a *knolle* deletion mutant, although the latter accumulated at the cell plate, whereas the former was detected at the PVC. In contrast, SYP112 of the plasma-membrane group, although normally dispensable, fully substituted for KNOLLE. Thus three factors contribute to syntaxin specificity in cytokinesis: strong expression during M phase, absence of a sorting sequence, and protein function during cell plate formation.

ENDOCYTOSIS AND LOCAL MEMBRANE RECYCLING The cell plate shrinks in surface area and volume by approximately 70% during consolidation, presumably by clathrin-mediated endocytosis (Otegui et al. 2001). During cell plate expansion, KNOLLE disappeared from the center and was concentrated at the margin (Lauber et al. 1997). Evidence for local membrane recycling is scant at present. Brefeldin A treatment of *Arabidopsis* dividing root cells caused accumulation of KNOLLE and PIN1 in the same BFA compartments (Geldner et al. 2001). In dividing cells expressing BFA-resistant GNOM ARF-GEF, PIN1 no longer accumulated in BFA compartments but KNOLLE did, suggesting that internalized proteins are sorted at endosomes (Geldner et al. 2003). KNOLLE was proposed to accumulate at the margin of the expanding cell plate by local recycling via endosomes (Jürgens & Pacher 2003). It is conceivable that lateral diffusion of proteins within the cell plate is hindered by its lipid composition, which may necessitate membrane retrieval and local recycling. A similar argument has been put forward for the maintenance of the asymmetric localization of plasma-membrane proteins in *Arabidopsis* and for the maintenance of cell polarity in yeast (Jürgens & Geldner 2002, Valdez-Taubas & Pelham 2003).

FUTURE PERSPECTIVES

The analysis of membrane trafficking in plants is coming of age. The increasing use of GFP marker technology in conjunction with transient expression assays in cultured cells has led to remarkable progress in the past several years. However, the current use of different cell types has its limitations. It is not obvious to what extent findings in one system can be transferred to a different system, which is illustrated by the differential effects of BFA. Another concern is the use of heterologous markers and transient expression assays, which may not reflect the physiological situation in the intact organism. These problems could be overcome by studying trafficking processes in the genetic model *Arabidopsis*. This should also resolve the current conflict about vacuolar sorting receptors.

The results obtained so far support the view that many aspects of membrane trafficking are conserved between plants and nonplant organisms. However, both the endomembrane system and trafficking pathways have plant-specific features that cannot be studied in nonplant organisms. These include not only trafficking to the storage vacuole or during cytokinesis but also rapid recycling between plasma membrane and endosomes. Considering the current rate of progress, it

may not take long until our understanding of membrane trafficking in plants has reached a comparable level of sophistication as in yeast or animals.

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LITERATURE CITED

Ahmed SU, Rojo E, Kovaleva V, Venkataraman S, Dombrowski JE, et al. 2000. The plant vacuolar sorting receptor AtELP is involved in transport of NH_2-terminal propeptide-containing vacuolar proteins in *Arabidopsis thaliana. J. Cell Biol.* 149:1335–44

Assaad FF, Huet Y, Mayer U, Jürgens G. 2001. The cytokinesis gene KEULE encodes a Sec1 protein that binds the syntaxin KNOLLE. *J. Cell Biol.* 152:531–43

Baluska F, Hlavacka A, Samaj J, Palme K, Robinson DG, et al. 2002. F-actin-dependent endocytosis of cell wall pectins in meristematic root cells. Insights from brefeldin A-induced compartments. *Plant Physiol.* 130: 422–31

Barlowe C. Molecular recognition of cargo by the COPII complex: a most accommodating coat. 2003. *Cell* 114:395–97

Bar-Peled M, Raikhel NV. 1997. Characterization of AtSEC12 and AtSAR1. Proteins likely involved in endoplasmic reticulum and Golgi transport. *Plant Physiol.* 114:315–24

Barth M, Holstein SEH. 2004. Identification and functional characterization of *Arabidopsis* AP180, a binding partner of plant αC-adaptin. *J. Cell Sci.* 117:2051–62

Bassham DC, Raikhel NV. 1999. The pre-vacuolar t-SNARE AtPEP12p forms a 20S complex that dissociates in the presence of ATP. *Plant J.* 19:599–603

Bassham DC, Sanderfoot AA, Kovaleva V, Zheng H, Raikhel NV. 2000. AtVPS45 complex formation at the *trans*-Golgi network. *Mol. Biol. Cell.* 11:2251–65

Batoko H, Zheng HQ, Hawes C, Moore I. 2000. A rab1 GTPase is required for transport between the endoplasmic reticulum and Golgi apparatus and for normal Golgi movement in plants. *Plant Cell* 12:2201–18

Battey NH, James NC, Greenland AJ, Brownlee C. Exocytosis and endocytosis. 1999. *Plant Cell* 11:643–60

Benghezal M, Wasteneys GO, Jones DA. 2000. The C-terminal dilysine motif confers endoplasmic reticulum localization to type I membrane proteins in plants. *Plant Cell* 12:1179–201

Bigay J, Gounon P, Robineau S, Antonny B. 2003. Lipid packing sensed by ArfGAP1 couples COPI coat disassembly to membrane bilayer curvature. *Nature* 426:563–66

Boehm M, Bonifacino JS. 2001. Adaptins: the final recount. *Mol. Biol. Cell.* 12:2907–20

Boevink P, Oparka K, Santa Cruz S, Martin B, Betteridge A, Hawes C. 1998. Stacks on tracks: the plant Golgi apparatus traffics on an actin/ER network. *Plant J.* 15:441–47

Boisson M, Gomord V, Audran C, Berger N, Dubreucq B, et al. 2001. *Arabidopsis* glucosidase I mutants reveal a critical role of N-glycan trimming in seed development. *EMBO J.* 20:1010–19

Bolte S, Brown S, Satiat-Jeunemaitre B. 2004. The N-myristoylated Rab-GTPase m-Rab_{mc} is involved in post-Golgi trafficking events to the lytic vacuole in plant cells. *J. Cell Sci.* 117:943–54

Bonifacino JS, Lippincott-Schwartz J. 2003. Coat proteins: shaping membrane transport. *Nat. Rev. Mol. Cell Biol.* 4:409–14

Bonifacino JS, Traub LM. 2003. Signals for sorting of transmembrane proteins to endosomes and lysosomes. *Annu. Rev. Biochem.* 72:395–447

Brandizzi F, Fricker M, Hawes C. 2002a. A greener world: the revolution in plant

bioimaging. *Nat. Rev. Mol. Cell Biol.* 3:520–30

Brandizzi F, Hanton S, DaSilva LL, Boevink P, Evans D, et al. 2003. ER quality control can lead to retrograde transport from the ER lumen to the cytosol and the nucleoplasm in plants. *Plant J.* 34:269–81

Brandizzi F, Snapp EL, Roberts AG, Lippincott-Schwartz J, Hawes C. 2002b. Membrane protein transport between the endoplasmic reticulum and the Golgi in tobacco leaves is energy dependent but cytoskeleton independent: evidence from selective photobleaching. *Plant Cell* 14:1293–309

Cao X, Rogers SW, Butler J, Beevers L, Rogers JC. 2000. Structural requirements for ligand binding by a probable plant vacuolar sorting receptor. *Plant Cell* 12:493–506

Chrispeels MJ, Herman EM. 2000. Endoplasmic reticulum-derived compartments function in storage and as mediators of vacuolar remodeling via a new type of organelle, precursor protease vesicles. *Plant Physiol.* 123:1227–34

Clague MJ, Urbe S. 2001. The interface of receptor trafficking and signalling. *J. Cell Sci.* 114:3075–81

Collins NC, Thordal-Christensen H, Lipka V, Bau S, Kombrink E, et al. 2003. SNARE-protein-mediated disease resistance at the plant cell wall. *Nature* 425:973–77

Crofts AJ, Leborgne-Castel N, Hillmer S, Robinson DG, Phillipson B, et al. 1999. Saturation of the endoplasmic reticulum retention machinery reveals anterograde bulk flow. *Plant Cell* 11:2233–48

Cutler SR, Ehrhardt DW. 2002. Polarized cytokinesis in vacuolate cells of *Arabidopsis*. *Proc. Natl. Acad. Sci. USA* 99:2812–17

Danino D, Hinshaw JE. 2001. Dynamin family of mechanoenzymes. *Curr. Opin. Cell Biol.* 13:454–60

da Silva Conceicao A, Marty-Mazars D, Bassham DC, Sanderfoot AA, Marty F, Raikhel NV. 1997. The syntaxin homolog AtPEP12p resides on a late post-Golgi compartment in plants. *Plant Cell* 9:571–82

d'Enfert C, Gensse M, Gaillardin C. 1992. Fission yeast and a plant have functional homologues of the Sar1 and Sec12 proteins involved in ER to Golgi traffic in budding yeast. *EMBO J.* 11:4205–11

Di Sansebastiano GP, Paris N, Marc-Martin S, Neuhaus JM. 2001. Regeneration of a lytic central vacuole and of neutral peripheral vacuoles can be visualized by green fluorescent proteins targeted to either type of vacuoles. *Plant Physiol.* 126:78–86

Dupree P, Sherrier DJ. 1998. The plant Golgi apparatus. *Biochim. Biophys. Acta* 1404: 259–70

Elias M, Drdova E, Ziak D, Bavlnka B, Hala M, et al. 2003. The exocyst complex in plants. *Cell Biol. Int.* 27:199–20

Emans N, Zimmermann S, Fischer R. 2002. Uptake of a fluorescent marker in plant cells is sensitive to brefeldin A and wortmannin. *Plant Cell* 14:71–86

Filippini F, Rossi V, Galli T, Budillon A, D'Urso M, D'Esposito M. 2001. Longins: a new evolutionary conserved VAMP family sharing a novel SNARE domain. *Trends Biochem. Sci.* 26:407–9

Geelen D, Leyman B, Batoko H, Di Sansebastiano GP, Moore I, et al. 2002. The abscisic acid-related SNARE homolog NtSyr1 contributes to secretion and growth: evidence from competition with its cytosolic domain. *Plant Cell* 14:387–406

Geldner N, Anders N, Wolters H, Keicher J, Kornberger W, et al. 2003. The *Arabidopsis* GNOM ARF-GEF mediates endosomal recycling, auxin transport, and auxin-dependent plant growth. *Cell* 112:219–30

Geldner N, Friml J, Stierhof YD, Jürgens G, Palme K. 2001. Auxin transport inhibitors block PIN1 cycling and vesicle trafficking. *Nature* 413:425–28

Gillmor CS, Poindexter P, Lorieau J, Palcic MM, Somerville C. 2002. Alpha-glucosidase I is required for cellulose biosynthesis and morphogenesis in *Arabidopsis*. *J. Cell Biol.* 156:1003–13

Grebe M, Xu J, Mobius W, Ueda T, Nakano A, et al. 2003. *Arabidopsis* sterol endocytosis involves actin-mediated trafficking via

ARA6-positive early endosomes. *Curr. Biol.* 13:1378–87

Gruenberg J. The endocytic pathway: a mosaic of domains. 2001. *Nat. Rev. Mol. Cell Biol.* 2:721–30

Hadlington JL, Denecke J. 2000. Sorting of soluble proteins in the secretory pathway of plants. *Curr. Opin. Plant Biol.* 3:461–68

Happel N, Honing S, Neuhaus JM, Paris N, Robinson DG, Holstein SE. 2004. *Arabidopsis* micro A-adaptin interacts with the tyrosine motif of the vacuolar sorting receptor VSR-PS1. *Plant J.* 37:678–93

Hara-Nishimura II, Shimada T, Hatano K, Takeuchi Y, Nishimura M. 1998. Transport of storage proteins to protein storage vacuoles is mediated by large precursor-accumulating vesicles. *Plant Cell* 10:825–36

Heese M, Gansel X, Sticher L, Wick P, Grebe M, et al. 2001. Functional characterization of the KNOLLE-interacting t-SNARE At-SNAP33 and its role in plant cytokinesis. *J. Cell Biol.* 155:239–49

Hillmer S, Movafeghi A, Robinson DG, Hinz G. 2001. Vacuolar storage proteins are sorted in the *cis*-cisternae of the pea cotyledon Golgi apparatus. *J. Cell Biol.* 152:41–50

Hinz G, Hillmer S, Baumer M, Hohl I. 1999. Vacuolar storage proteins and the putative vacuolar sorting receptor BP-80 exit the Golgi apparatus of developing pea cotyledons in different transport vesicles. *Plant Cell* 11:1509–24

Hoh B, Hinz G, Jeong BK, Robinson DG. 1995. Protein storage vacuoles form de novo during pea cotyledon development. *J. Cell Sci.* 108:299–310

Hohl I, Robinson DG, Chrispeels MJ, Hinz G. 1996. Transport of storage proteins to the vacuole is mediated by vesicles without a clathrin coat. *J. Cell Sci.* 109:2539–50

Holstein SE. 2002. Clathrin and plant endocytosis. *Traffic* 3:614–20

Hong Z, Bednarek SY, Blumwald E, Hwang I, Jürgens G, et al. 2003. A unified nomenclature for *Arabidopsis* dynamin-related large GTPases based on homology and possible functions. *Plant Mol. Biol.* 53:261–65

Hong Z, Delauney AJ, Verma DP. 2001a. A cell plate-specific callose synthase and its interaction with phragmoplastin. *Plant Cell* 13: 755–68

Hong Z, Zhang Z, Olson JM, Verma DP. 2001b. A novel UDP-glucose transferase is part of the callose synthase complex and interacts with phragmoplastin at the forming cell plate. *Plant Cell* 13:769–79

Inaba T, Nagano Y, Nagasaki T, Sasaki Y. 2002. Distinct localization of two closely related Ypt3/Rab11 proteins on the trafficking pathway in higher plants. *J. Biol. Chem.* 277:9183–88

Irons SL, Evans DE, Brandizzi F. 2003. The first 238 amino acids of the human lamin B receptor are targeted to the nuclear envelope in plants. *J. Exp. Bot.* 54:943–50

Jackson CL, Casanova JE. 2000. Turning on ARF: the Sec7 family of guanine-nucleotide-exchange factors. *Trends Cell Biol.* 10:60–67

Jackson MR, Nilsson T, Peterson PA. 1993. Retrieval of transmembrane proteins to the endoplasmic reticulum. *J. Cell Biol.* 121:317–33

Jahn R, Lang T, Südhof TC. 2003. Membrane fusion. *Cell* 112:519–33

Jauh GY, Fischer AM, Grimes HD, Ryan CA Jr, Rogers JC. 1998. delta-Tonoplast intrinsic protein defines unique plant vacuole functions. *Proc. Natl. Acad. Sci. USA* 95:12995–99

Jiang L, Phillips TE, Hamm CA, Drozdowicz YM, Rea PA, et al. 2001. The protein storage vacuole: a unique compound organelle. *J. Cell Biol.* 155:991–1002

Jiang L, Phillips TE, Rogers SW, Rogers JC. 2000. Biogenesis of the protein storage vacuole crystalloid. *J. Cell Biol.* 150:755–70

Jin JB, Kim YA, Kim SJ, Lee SH, Kim DH, et al. 2001. A new dynamin-like protein, ADL6, is involved in trafficking from the *trans*-Golgi network to the central vacuole in *Arabidopsis. Plant Cell* 13:1511–26

Jürgens G, Geldner N. 2002. Protein secretion in plants: from the *trans*-Golgi network to the outer space. *Traffic* 3:605–13

Jürgens G, Pacher T. 2003. Cytokinesis: membrane trafficking by default? *Annu. Plant Rev.* 9:238–54

Kang BH, Busse JS, Bednarek SY. 2003. Members of the *Arabidopsis* dynamin-like gene family, ADL1, are essential for plant cytokinesis and polarized cell growth. *Plant Cell* 15:899–913

Kang BH, Busse JS, Dickey C, Rancour DM, Bednarek SY. 2001. The *Arabidopsis* cell plate-associated dynamin-like protein, ADL1Ap, is required for multiple stages of plant growth and development. *Plant Physiol.* 126:47–68

Kim DH, Eu YJ, Yoo CM, Kim YW, Pih KT, et al. 2001. Trafficking of phosphatidylinositol 3-phosphate from the *trans*-Golgi network to the lumen of the central vacuole in plant cells. *Plant Cell* 13:287–301

Kirsch T, Saalbach G, Raikhel NV, Beevers L. 1996. Interaction of a potential vacuolar targeting receptor with amino- and carboxyl-terminal targeting determinants. *Plant Physiol.* 111:469–74

Lam BC, Sage TL, Bianchi F, Blumwald E. 2002. Regulation of ADL6 activity by its associated molecular network. *Plant J.* 31:565–76

Lauber MH, Waizenegger I, Steinmann T, Schwarz H, Mayer U, et al. 1997. The *Arabidopsis* KNOLLE protein is a cytokinesis-specific syntaxin. *J. Cell Biol.* 139:1485–93

Lee MH, Min MK, Lee YJ, Jin JB, Shin DH, et al. 2002. ADP-ribosylation factor 1 of *Arabidopsis* plays a critical role in intracellular trafficking and maintenance of endoplasmic reticulum morphology in *Arabidopsis. Plant Physiol.* 129:1507–20

Lenhard M, Laux T. 2003. Stem cell homeostasis in the *Arabidopsis* shoot meristem is regulated by intercellular movement of CLAVATA3 and its sequestration by CLAVATA1. *Development* 130:3163–73

Leyman B, Geelen D, Blatt MR. 2000. Localization and control of expression of Nt-Syr1, a tobacco SNARE protein. *Plant J.* 24:369–81

Low SH, Li X, Miura M, Kudo N, Quinones B, Weimbs T. 2003. Syntaxin 2 and endobrevin are required for the terminal step of cytokinesis in mammalian cells. *Dev. Cell* 4:753–59

Lukowitz W, Nickle TC, Meinke DW, Last RL, Conklin PL, Somerville CR. 2001. Arabidopsis *cyt1* mutants are deficient in a mannose-1-phosphate guanylyltransferase and point to a requirement of N-linked glycosylation for cellulose biosynthesis. *Proc. Natl. Acad. Sci. USA* 98:2262–67

Marty F. 1999. Vacuoles. *Plant Cell* 11:587–99

Matsubayashi Y. 2003. Ligand-receptor pairs in plant peptide signaling. *J. Cell Sci.* 116: 3863–70

Matsuoka K, Bassham DC, Raikhel NV, Nakamura K. 1995. Different sensitivity to wortmannin of two vacuolar sorting signals indicates the presence of distinct sorting machineries in tobacco cells. *J. Cell Biol.* 130:1307–18

Matsuoka K, Neuhaus JM. 1999. *Cis*-elements of protein transport to the plant vacuoles. *J. Exp. Bot.* 50:165–174

Movafeghi A, Happel N, Pimpl P, Tai GH, Robinson DG. 1999. *Arabidopsis* Sec21p and Sec23p homologs. Probable coat proteins of plant COP-coated vesicles. *Plant Physiol.* 119:1437–46

Müller I, Wagner W, Völker A, Schellmann S, Nacry P, et al. 2003. Syntaxin specificity of cytokinesis in *Arabidopsis. Nat. Cell Biol.* 5: 531–34

Murshid A, Presley JF. 2004. ER-to-Golgi transport and cytoskeletal interactions in animal cells. *Cell. Mol. Life Sci.* 61:133–45

Nahm MY, Kim SW, Yun D, Lee SY, Cho MJ, Bahk JD. 2003. Molecular and biochemical analyses of OsRab7, a rice Rab7 homolog. *Plant Cell Physiol.* 44:1341–49

Nebenführ A, Staehelin LA. 2001. Mobile factories: Golgi dynamics in plant cells. *Trends Plant Sci.* 6:160–67

Neumann U, Brandizzi F, Hawes C. 2003. Protein transport in plant cells: in and out of the Golgi. *Ann. Bot.* 92:167–80

Nie Z, Boehm M, Boja ES, Vass WC, Bonifacino JS, et al. 2003. Specific regulation of

the adaptor protein complex AP-3 by the Arf GAP AGAP1. *Dev. Cell* 5:513–21

Nühse TS, Boller T, Peck SC. 2003. A plasma membrane syntaxin is phosphorylated in response to the bacterial elicitor flagellin. *J. Biol. Chem.* 278:45248–54

Otegui MS, Mastronarde DN, Kang BH, Bednarek SY, Staehelin LA. 2001. Three-dimensional analysis of syncytial-type cell plates during endosperm cellularization visualized by high resolution electron tomography. *Plant Cell* 13:2033–51

Otegui MS, Staehelin LA. 2004. Electron tomographic analysis of post-meiotic cytokinesis during pollen development in *Arabidopsis thaliana*. *Planta* 218:501–15

Paris N, Rogers SW, Jiang L, Kirsch T, Beevers L, et al. 1997. Molecular cloning and further characterization of a probable plant vacuolar sorting receptor. *Plant Physiol.* 115:29–39

Paris N, Stanley CM, Jones RL, Rogers JC. 1996. Plant cells contain two functionally distinct vacuolar compartments. *Cell* 85: 563–72

Park M, Kim SJ, Vitale A, Hwang I. 2004. Identification of the protein storage vacuole and protein targeting to the vacuole in leaf cells of three plant species. *Plant Physiol.* 134:625–39

Peterson MR, Emr SD. 2001. The class C Vps complex functions at multiple stages of the vacuolar transport pathway. *Traffic* 2:476–86

Pfeffer SR. 2001. Membrane transport: retromer to the rescue. *Curr. Biol.* 11:R109–11

Phillipson BA, Pimpl P, daSilva LL, Crofts AJ, Taylor JP, et al. 2001. Secretory bulk flow of soluble proteins is efficient and COPII dependent. *Plant Cell* 13:2005–20

Pimpl P, Hanton SL, Taylor JP, Pinto-daSilva LL, Denecke J. 2003. The GTPase ARF1p controls the sequence-specific vacuolar sorting route to the lytic vacuole. *Plant Cell* 15:1242–56

Pimpl P, Movafeghi A, Coughlan S, Denecke J, Hillmer S, Robinson DG. 2000. In situ localization and in vitro induction of plant COPI-coated vesicles. *Plant Cell* 12:2219–36

Raiborg C, Rusten TE, Stenmark H. 2003. Protein sorting into multivesicular endosomes. *Curr. Opin. Cell Biol.* 15:446–55

Rancour DM, Dickey CE, Park S, Bednarek SY. 2002. Characterization of AtCDC48. Evidence for multiple membrane fusion mechanisms at the plane of cell division in plants. *Plant Physiol.* 130:1241–53

Ritzenthaler C, Nebenfuhr A, Movafeghi A, Stussi-Garaud C, Behnia L, et al. 2002. Reevaluation of the effects of brefeldin A on plant cells using tobacco Bright Yellow 2 cells expressing Golgi-targeted green fluorescent protein and COPI antisera. *Plant Cell* 14:237–61

Rojo E, Gillmor CS, Kovaleva V, Somerville CR, Raikhel NV. 2001. *VACUOLELESS1* is an essential gene required for vacuole formation and morphogenesis in *Arabidopsis. Dev. Cell* 1:303–10

Rojo E, Sharma VK, Kovaleva V, Raikhel NV, Fletcher JC. 2002. CLV3 is localized to the extracellular space, where it activates the *Arabidopsis* CLAVATA stem cell signaling pathway. *Plant Cell* 14:969–77

Rojo E, Zouhar J, Kovaleva V, Hong S, Raikhel NV. 2003. The AtC-VPS protein complex is localized to the tonoplast and the prevacuolar compartment in *Arabidopsis. Mol. Biol. Cell* 14:361–69

Rutherford S, Moore I. 2002. The *Arabidopsis* Rab GTPase family: another enigma variation. *Curr. Opin. Plant Biol.* 5:518–28

Saint-Jore CM, Evins J, Batoko H, Brandizzi F, Moore I, Hawes C. 2002. Redistribution of membrane proteins between the Golgi apparatus and endoplasmic reticulum in plants is reversible and not dependent on cytoskeletal networks. *Plant J.* 29:661–78

Saint-Jore-Dupas C, Gomord V, Paris N. 2004. Protein localization in the plant Golgi apparatus and the *trans*-Golgi network. *Cell. Mol. Life Sci.* 61:159–71

Samuels AL, Giddings TH Jr., Staehelin LA. 1995. Cytokinesis in tobacco BY-2 and root tip cells: a new model of cell plate formation in higher plants. *J. Cell Biol.* 130:1345–57

Sanderfoot AA, Ahmed SU, Marty-Mazars D, Rapoport I, Kirchhausen T, et al. 1998. A

putative vacuolar cargo receptor partially colocalizes with AtPEP12p on a prevacuolar compartment in *Arabidopsis* roots. *Proc. Natl. Acad. Sci. USA* 95:9920–25

Sanderfoot AA, Assaad FF, Raikhel NV. 2000. The *Arabidopsis* genome. An abundance of soluble *N*-ethylmaleimide-sensitive factor adaptor protein receptors. *Plant Physiol.* 124:1558–69

Sanderfoot AA, Kovaleva V, Bassham DC, Raikhel NV. 2001a. Interactions between syntaxins identify at least five SNARE complexes within the Golgi/prevacuolar system of the *Arabidopsis*cell. *Mol. Biol. Cell* 12: 3733–43

Sanderfoot AA, Pilgrim M, Adam L, Raikhel NV. 2001b. Disruption of individual members of *Arabidopsis* syntaxin gene families indicates each has essential functions. *Plant Cell* 13:659–66

Sanderfoot AA, Raikhel NV. 2003. The secretory system of *Arabidopsis.* In *The Arabidopsis Book*, eds. CR Somerville, EM Meyerowitz. 24 pp. Rockville, MD: Am. Soc. Plant Biologists. doi/10.1199/tab.0098, http://www.aspb.org/publications/arabidopsis/

Sato K, Sato M, Nakano A. 2001. Rer1p, a retrieval receptor for endoplasmic reticulum membrane proteins, is dynamically localized to the Golgi apparatus by coatomer. *J. Cell Biol.* 152:935–44

Sato K, Ueda T, Nakano A. 1999. The *Arabidopsis thaliana RER1* gene family: its potential role in the endoplasmic reticulum localization of membrane proteins. *Plant Mol. Biol.* 41:815–24

Shah K, Russinova E, Gadella TW Jr, Willemse J, De Vries SC. 2002. The *Arabidopsis* kinase-associated protein phosphatase controls internalization of the somatic embryogenesis receptor kinase 1. *Genes Dev.* 16:1707–20

Shimada T, Fuji K, Tamura K, Kondo M, Nishimura M, Hara-Nishimura I. 2003. Vacuolar sorting receptor for seed storage proteins in *Arabidopsis thaliana*. *Proc. Natl. Acad. Sci. USA* 100:16095–100

Shimada T, Watanabe E, Tamura K, Hayashi Y, Nishimura M, Hara-Nishimura I. 2002. A vacuolar sorting receptor PV72 on the membrane of vesicles that accumulate precursors of seed storage proteins (PAC vesicles). *Plant Cell Physiol.* 43:1086–95

Sohn EJ, Kim ES, Zhao M, Kim SJ, Kim H, et al. 2003. Rha1, an *Arabidopsis* Rab5 homolog, plays a critical role in the vacuolar trafficking of soluble cargo proteins. *Plant Cell* 15:1057–70

Staehelin LA. 1997. The plant ER: a dynamic organelle composed of a large number of discrete functional domains. *Plant J.* 11:1151–65

Staehelin LA, Hepler PK. 1996. Cytokinesis in higher plants. *Cell* 84:821–24

Staehelin LA, Moore I. 1995. The plant Golgi apparatus: structure, functional organization and trafficking mechanisms. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* 46:261–88

Steinmann T, Geldner N, Grebe M, Mangold S, Jackson CL, et al. 1999. Coordinated polar localization of auxin efflux carrier PIN1 by GNOM ARF GEF. *Science* 286:316–18

Surpin M, Zheng H, Morita MT, Saito C, Avila E, et al. 2003. The VTI family of SNARE proteins is necessary for plant viability and mediates different protein transport pathways. *Plant Cell* 15:2885–99

Takeuchi M, Ueda T, Sato K, Abe H, Nagata T, Nakano A. 2000. A dominant negative mutant of sar1 GTPase inhibits protein transport from the endoplasmic reticulum to the Golgi apparatus in tobacco and *Arabidopsis* cultured cells. *Plant J.* 23:517–25

Takeuchi M, Ueda T, Yahara N, Nakano A. 2002. Arf1 GTPase plays roles in the protein traffic between the endoplasmic reticulum and the Golgi apparatus in tobacco and *Arabidopsis* cultured cells. *Plant J.* 31:499–515

Tse YC, Mo B, Hillmer S, Zhao M, Lo SW, et al. 2004. Identification of multivesicular bodies as prevacuolar compartments in *Nicotiana tabacum* BY-2 cells. *Plant Cell* 16:672–93

Ueda T, Yamaguchi M, Uchimiya H, Nakano A. 2001. Ara6, a plant-unique novel type Rab GTPase, functions in the endocytic pathway

of *Arabidopsis thaliana*. *EMBO J.* 20:4730–41

Valdez-Taubas J, Pelham HR. 2003. Slow diffusion of proteins in the yeast plasma membrane allows polarity to be maintained by endocytic cycling. *Curr. Biol.* 13:1636–40

Vanoosthuyse V, Tichtinsky G, Dumas C, Gaude T, Cock JM. 2003. Interaction of calmodulin, a sorting nexin and kinase-associated protein phosphatase with the *Brassica oleracea* S locus receptor kinase. *Plant Physiol.* 133:919–29

Vernoud V, Horton AC, Yang Z, Nielsen E. 2003. Analysis of the small GTPase gene superfamily of *Arabidopsis*. *Plant Physiol.* 131:1191–208

Vitale A, Denecke J. 1999. The endoplasmic reticulum—gateway of the secretory pathway. *Plant Cell* 11:615–28

Waizenegger I, Lukowitz W, Assaad F, Schwarz H, Jürgens G, Mayer U. 2000. The *Arabidopsis KNOLLE* and *KEULE* genes interact to promote vesicle fusion during cytokinesis. *Curr. Biol.* 10:1371–74

Ward TH, Brandizzi F. 2004. Dynamics of proteins in Golgi membranes: comparisons between mammalian and plant cells highlighted by photobleaching techniques. *Cell. Mol. Life Sci.* 61:172–85

Watanabe E, Shimada T, Kuroyanagi M, Nishimura M, Hara-Nishimura I. 2002. Calcium-mediated association of a putative vacuolar sorting receptor PV72 with a propeptide of 2S albumin. *J. Biol. Chem.* 277:8708–15

Watanabe E, Shimada T, Tamura K, Matsushima R, Koumoto Y, et al. 2004. An ER-localized form of PV72, a seed-specific vacuolar sorting receptor, interferes with the transport of an NPIR-containing proteinase in *Arabidopsis* leaves. *Plant Cell Physiol.* 45:9–17

Whyte JR, Munro S. 2002. Vesicle tethering complexes in membrane traffic. *J. Cell Sci.* 115:2627–37

Woodman PG. 2000. Biogenesis of the sorting endosome: the role of Rab5. *Traffic* 1:695–701

Wurmser AE, Sato TK, Emr SD. 2000. New component of the vacuolar class C-Vps complex couples nucleotide exchange on the Ypt7 GTPase to SNARE-dependent docking and fusion. *J. Cell Biol.* 151:551–62

Yano D, Sato M, Saito C, Sato MH, Morita MT, Tasaka M. 2003. A SNARE complex containing SGR3/AtVAM3 and ZIG/VTI11 in gravity-sensing cells is important for *Arabidopsis* shoot gravitropism. *Proc. Natl. Acad. Sci. USA* 100:8589–94

Yokoyama R, Nishitani K. 2001. Endoxyloglucan transferase is localized both in the cell plate and in the secretory pathway destined for the apoplast in tobacco cells. *Plant Cell Physiol.* 42:292–300

Zheng H, Bednarek SY, Sanderfoot AA, Alonso J, Ecker JR, Raikhel NV. 2002. NPSN11 is a cell plate-associated SNARE protein that interacts with the syntaxin KNOLLE. *Plant Physiol.* 129:530–39

Zhu J, Gong Z, Zhang C, Song CP, Damsz B, et al. 2002. OSM1/SYP61: a syntaxin protein in *Arabidopsis* controls abscisic acid-mediated and non-abscisic acid-mediated responses to abiotic stress. *Plant Cell* 14: 3009–28

Zhu Y, Traub LM, Kornfeld S. 1998. ADP-ribosylation factor 1 transiently activates high-affinity adaptor protein complex AP-1 binding sites on Golgi membranes. *Mol. Biol. Cell* 9:1323–37

Zuo J, Niu QW, Nishizawa N, Wu Y, Kost B, Chua NH. 2000. KORRIGAN, an *Arabidopsis* endo-1,4-beta-glucanase, localizes to the cell plate by polarized targeting and is essential for cytokinesis. *Plant Cell* 12:1137–52