

DGARM/C10orf76/ARMH3 for Ceramide Transfer Zone at the Endoplasmic Reticulum–Distal Golgi Contacts

Contact
Volume 7: 1–9
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DOI: 10.1177/25152564241239443
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Abstract

Phosphatidylinositol 4-monophosphate (PtdIns(4)P) is one of the key membrane components which mark the membrane contact sites. In the mammalian Golgi complex, PtdIns(4)P is produced at various subregions via specific mechanisms for each site. Particularly, PtdIns(4)P pools generated at the distal Golgi regions are pivotal for the determination of membrane contacts between the endoplasmic reticulum (ER) and Golgi, at which inter-organelle lipid transport takes place. In this short review, we will focus on C10orf76 (or ARMH3), which we propose to rename as DGARM after a distal Golgi armadillo repeat protein, for its function in generating a PtdIns(4)P pool crucial for ER-to-distal Golgi ceramide transport. We further discuss from the viewpoint of the evolutionary conservation of DGARM.

Keywords

lipid transfer proteins, phosphatidylinositol 4-monophosphate, sphingomyelin, PI4KB, ACBD3, CERT

Introduction

Eukaryotic cells have various membrane-bound compartments called organelles. In addition, each organelle seemingly has various sub-regions (or zones) with different functions (Sasaki and Yoshida, 2019; Nishitoh, 2019). Membrane contact sites are the subcellular regions consisting of two different organelle membranes within ~30 nm distance and the cytosol fraction between them. Although organelle contacts have been recognized early in 1960s in cell images by classic electron microscopy, their functions have been clarified since 2000s. As a pivotal function of membrane contact sites, inter-organelle transport of lipids by various lipid transfer proteins (LTPs) has been extensively studied in this decade; providing mechanistic insights into the long enigmas of how cell properly accomplishes intracellular trafficking of lipids or of how a specific lipid type is picked up from a specific organelle and then delivered to another specific organelle in a cell (reviewed in Hanada, 2018; Balla et al., 2019; Wong et al., 2019). Phosphatidylinositol 4-monophosphate (PtdIns(4)P) is a well-studied membrane component for its role in specifying various contact sites. We recently found that C10orf76 (alias ARMH3), a PtdIns 4-kinase-binding protein, is recruited to the distal Golgi and plays a pivotal role in the ER-to-distal Golgi trafficking of ceramide for the synthesis of sphingomyelin (Mizuike et al., 2023). Together with the predicted structure of C10orf76 as an armadillo (ARM)-like fold (Figure 1), consisting of α -helices arranged into a characteristic

superhelix (Tewari et al., 2010), we proposed to rename it to DGARM after a distal Golgi armadillo repeat protein. In this short review, we will describe a DGARM-relevant PtdIns(4)P pool critical for determining the ER–distal Golgi contacts. Moreover, we will make a phylogenetic discussion on DGARM.

The Architecture of the Golgi complex

The Golgi complex is an organelle with a distinctive structure (Figure 2A and B): multiple cisternae stacking in a polarized manner. Golgi cisternae can be classified as *cis*, *medial*, or *trans*, with the *cis* side being proximal (or early) in the antero-grade membrane traffic starting from the endoplasmic reticulum (ER), and the *trans* side being distal (or late). Additionally, the ER-Golgi intermediate compartment (ERGIC) and *trans*-Golgi network (TGN) are also thought to be subregions of the Golgi

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Received December 13, 2023. Revised February 26, 2024. Accepted February 28, 2024.

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C10orf76: chromosome 10 orf 76

ARMH3: Armadillo-like helical domain-containing protein 3

DGARM: distal Golgi Armadillo repeat protein

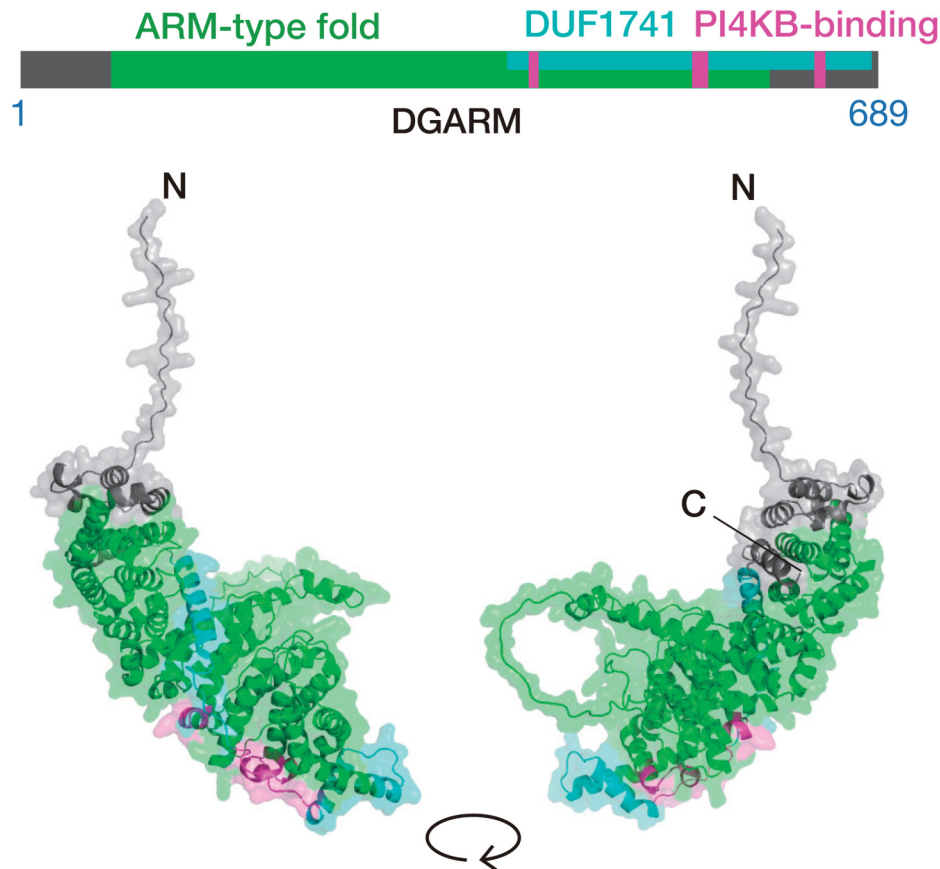


Figure 1. DGARM, a predicted armadillo-fold protein. A predicted structure of DGARM by alphafold (Jumper et al., 2021; Varadi et al., 2022; ver. 2022-11-01). ARM-type fold region (InterPro IPR016024) and regions involved in PI4KB-binding (McPhail et al., 2020) are colored green and magenta, respectively. DUF1741; a domain of unknown function, conserved only in primary sequences of DGARM orthologs. DUF1741 is colored cyan, but the colors of ARM-type fold or PI4KB-binding regions are prioritized.

complex (Pantazopoulou and Glick, 2019). Although the Golgi apparatus in several fungi, including the model yeast *Saccharomyces cerevisiae*, do not form a stacking structure under conventional growth conditions, the punctate cisternae are classified into *cis*-, *medial*-, and *trans*-Golgi (Suda and Nakano, 2012). The Golgi complex is regarded to be compartmentalized in two ways, separation of cisternae and intracisterna segregation (Tie et al., 2018); thereby allowing accurate sequential processing/modification of proteins and lipids. Particular lipids show heterogeneous distribution among the cisternae: for instance, sphingomyelin, cholesterol and PtdIns(4)P levels are higher at the distal Golgi than the proximal (Moreau and Cassagne, 1994; D'Angelo et al., 2008; Del Bel and Brill, 2018). These distribution gradients of lipids are generated by local lipid transport coupled with lipid metabolism.

PtdIns(4)P as a Determinant of the Lipid Transport Zones at the Distal Golgi

In the *de novo* synthetic pathway of sphingolipids, ceramide is synthesized in the ER and then transported to the Golgi complex for further conversion to obtain complex sphingolipids (Figure 2A). The major phosphosphingolipids, which we focus on in this review, varied in evolution (Figure 2A): sphingomyelin in vertebrates, ceramide phosphoethanolamine (CPE) in invertebrates, and phosphoinositol-containing sphingolipids in fungi and plants, while some invertebrates synthesize sphingomyelin (Panevska et al., 2019). Whereas ceramide trafficking for inositol phosphorylceramide (IPC) synthesis in the budding yeast *S. cerevisiae*, depends both on vesicular transport and LTPs Nvj2 and Svf1 (Kajiwara et al., 2014; Liu et al., 2017;

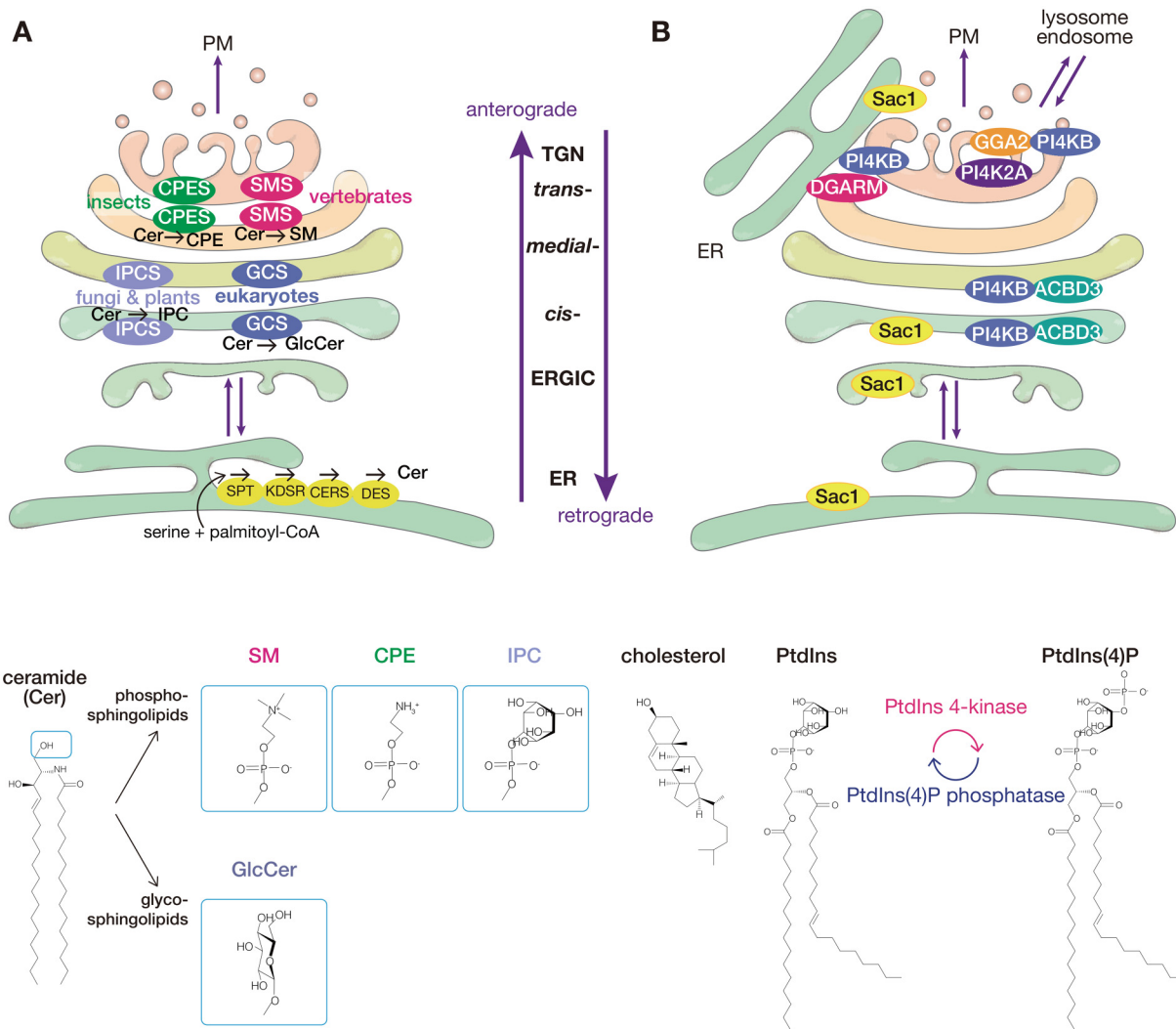


Figure 2. Distal Golgi–ER contact enables bypass of the proximal Golgi. (A) The intracellular distribution of ceramide-metabolizing enzymes in eukaryotes. For *de novo* ceramide synthesis machinery, mammalian enzymes are shown as a representative. (B) Golgi–PtdIns(4)P is generated by PI4KB or PI4K2A. While PI4K2A is palmitoylated at the TGN in a cholesterol-dependent manner for membrane association, PI4KB utilizes multiple recruitment systems to localize at various regions within the Golgi complex. PtdIns(4)P-phosphatase Sac1 cycles between the ER and the proximal Golgi, keeping the proximal PtdIns(4)P levels low. ER-resident Sac1 also catalyzes PtdIns(4)P turnover at the ER–TGN contacts. (Lower) Chemical structures of lipids discussed in this review. Only the polar headgroups are shown (enclosed in cyan boxes) for complex sphingolipids. Note that the major ceramide backbone in plants and fungi is phytoceramide, which has a hydroxyl group at the C4 position. SPT, serine-palmitoyltransferase; KDSR, 3-ketodihydrosphingosine reductase; CERS, ceramide synthase; DES, sphingolipid delta(4)-desaturase; IPCS, inositol phosphorylceramide synthase in fungi and plants; GCS, glucosylceramide synthase in eukaryotes; CPES, (insect-specific) CPE synthase; SMS, sphingomyelin synthase in vertebrates.

Limar et al., 2023), the synthesis of sphingomyelin and CPE highly depends on an LTP, the ceramide transport protein CERT. CERT and oxysterol-binding protein (OSBP) are representative LTPs working at the Golgi complex for ER-to-Golgi transport of ceramide and cholesterol, respectively (Hanada et al., 2003; Mesmin et al., 2013). These LTPs share similar domain construction: a pleckstrin homology (PH) domain which binds PtdIns(4)P, a “two phenylalanines in an acidic tract” (FFAT) motif that binds ER-resident protein VAPs, and a lipid transfer domain (i.e., a steroidogenic acute regulatory protein-related lipid transfer [START] domain in CERT

and an OSBP-related domain [ORD] in OSBP; Goto et al., 2020). Dual targeting via interactions between PH–PtdIns(4)P and FFAT–VAP enables these LTPs to localize at the ER–Golgi contacts. A previous study suggested that OSBP displays only a weak or indirect interaction with CERT in CHO cells (Perry and Ridgway, 2006), and a recent study showed that TMED2/10 form a supercomplex with OSBP, CERT, VAP-A in a PtdIns(4)P-, and ARF1-dependent manner in HeLa cells (Anwar et al., 2022). Thus, a partial, even if not all, population of Golgi-bound CERT, may act with OSBP at the same contact sites.

Reconstruction of the 3D Golgi structure from electron microscope tomography showed that specific regions of the ER adhere with the *trans*-cisternae or TGN (Mogelsvang et al., 2004; Venditti et al., 2019), suggesting that the ER–Golgi contacts are mainly formed at the distal Golgi in mammalian cells. Within the Golgi complex, the site for *de novo* synthesis of sphingomyelin is more distal than the site for the synthesis of glucosylceramide (Halter et al., 2007; Huitema et al., 2004; D’Angelo et al., 2007; Hayashi et al., 2018). The enrichment of PtdIns(4)P at the distal Golgi leads CERT and OSBP to preferentially localize at this region. This enables an efficient influx of ceramide to the site of sphingomyelin synthesis, which should provide a rationale for the intra-Golgi gradient of sphingomyelin. However, it is not clear whether PtdIns(4)P is produced at contact sites or PtdIns(4)P production induces formation of organelle contacts. Meanwhile, both could happen; PtdIns(4)P could be necessary for contacts to form but might also be needed to be produced more to maintain contacts. Note that the ability of OSBP to counter-transport cholesterol and PtdIns(4)P confers PtdIns(4)P to serve as the “fuel” to drive the ER-to-Golgi uphill movement of cholesterol by coupling with the Golgi-to-ER downhill movement of PtdIns(4)P (de Saint-Jean et al., 2011; Mesmin et al., 2013).

PtdIns(4)P synthesis in the Golgi complex principally depends on two PtdIns 4-kinases, PI4KB and PI4K2A (also known as PI4KIII β and PI4KII α , respectively; Figure 2B). PI4K2A is palmitoylated at the TGN in a cholesterol-dependent manner and is regarded to be localized at cholesterol-rich membrane domains in the TGN and endosomes (Balla et al., 2002; Lu et al., 2012). On the other hand, PI4KB is mostly dispersed in the cytosol and only a portion is recruited to the Golgi membrane (Blomen et al., 2015; McPhail et al., 2020). PI4KB adopts several types of Golgi-localization machinery in mammalian cells; that is, via interaction with ACBD3, DGARM (or C10orf76, ARMH3), or GGA2 (Sasaki et al., 2012; Mizuike et al., 2023; Daboussi et al., 2017). This allows PI4KB to localize at various regions within the Golgi complex (Figure 2B): since ACBD3 predominantly localizes at the proximal regions (Tie et al., 2018; Mizuike et al., 2023), DGARM at the distal regions (Mizuike et al., 2023), and GGA2 at the TGN (Doray et al., 2002). Although further observation of PI4KB using a super-resolution microscopy is demanded to prove this model. These PI4KB-binding proteins further interact with other proteins and will hence establish characteristic and functional differences in PtdIns(4)P pools. Amongst these, DGARM was shown to interact with GBF1 (Chan et al., 2019), which likely leads to the activation of PI4KB by ARF1, allowing vigorous PtdIns(4)P production at the distal Golgi.

A pool of PtdIns(4)P generated by the DGARM–PI4KB axis was shown to be important for defining the site at which CERT works to provide ceramide for sphingomyelin synthesis (Mizuike et al., 2023), as discussed above. In this study, ACBD3 was also indicated to partially compensate

for this function. In addition, DGARM–PI4KB axis-dependent PtdIns(4)P production and the downstream lipid transport were suggested to be critical for the activation of the cGAS–STING pathway, a DNA-triggered innate immunity system (Fang et al., 2023). STING translocates from the ER to the Golgi upon ligand binding and is palmitoylated and phosphorylated for activation (Mukai et al., 2016; Zhang et al., 2019; Ritchie et al., 2022). STING–DGARM interaction enables the local PtdIns(4)P production for recruiting LTPs (mainly CERT and ORPs including OSBP), thereby fine-tuning the environment of distal Golgi membrane into that suitable for STING activation. In this connection, ACBD3 was reported to be involved in the upstream step of STING activation: the ER-to-*cis*-Golgi transport of STING (Motani et al., 2022). ACBD3 concentrates ligand-bound STING in the face of ERGIC or *cis*-Golgi cisternae for efficient export from the ER, although PI4KB may be dispensable in this process. Still, it is of interest that PI4KB-binding proteins work sequentially in the same pathway at different regions.

Phylogenetic Relevance of PtdIns(4)P Production Machinery at the Distal Golgi

PtdIns(4)P is an essential lipid for eukaryotic life (Dickson and Hille, 2019; Posor et al., 2022), and PI4KB orthologs are widely conserved throughout eukaryotes (Figure 3A). Concurrently, ARF1 and GBF1, involved in PI4KB activation, are also widely conserved. However, genes involved in recruiting PI4KB to the Golgi complex are not so widely conserved (Figure 3A). ACBD3 is conserved only in *Holozoa* while DGARM is more widely conserved across the eukaryotes including fungi, plants and protists (Figure 3A and B), suggesting that the DGARM–PI4KB axis occurred earlier than the ACBD3–PI4KB axis in the evolution of life (Chan et al., 2019).

Nonetheless, DGARM was lost in several species including major model organisms *Caenorhabditis elegans*, *S. cerevisiae*, and *Arabidopsis thaliana*. FASTA search (<https://www.genome.jp/tools/fasta/>) for DGARM (seq. ID hsa:79591) suggested that DGARM is highly conserved in *Holozoa*, however, six of seven nematode species (registered in KEGG GENES) have lost DGARM. Notably, the PI4KB homolog in *Trichinella spiralis* (DGARM-positive) possesses the N-lobe kinase linker, which was suggested to contain the DGARM-binding region (McPhail et al., 2020), while that in *C. elegans* (DGARM-lost) does not (Figure 3C). For a detailed review of the DGARM-binding region of PI4KB, see (McPhail and Burke, 2023). It is conceivable that organisms which had lost DGARM have developed other (unique) systems for recruiting PI4KB orthologs to the distal or late Golgi regions. For instance, *S. cerevisiae* utilizes Frq1–Arf1 for recruiting Pik1, the PI4KB homolog in fungi, to the late Golgi (Strahl et al., 2005; Highland and

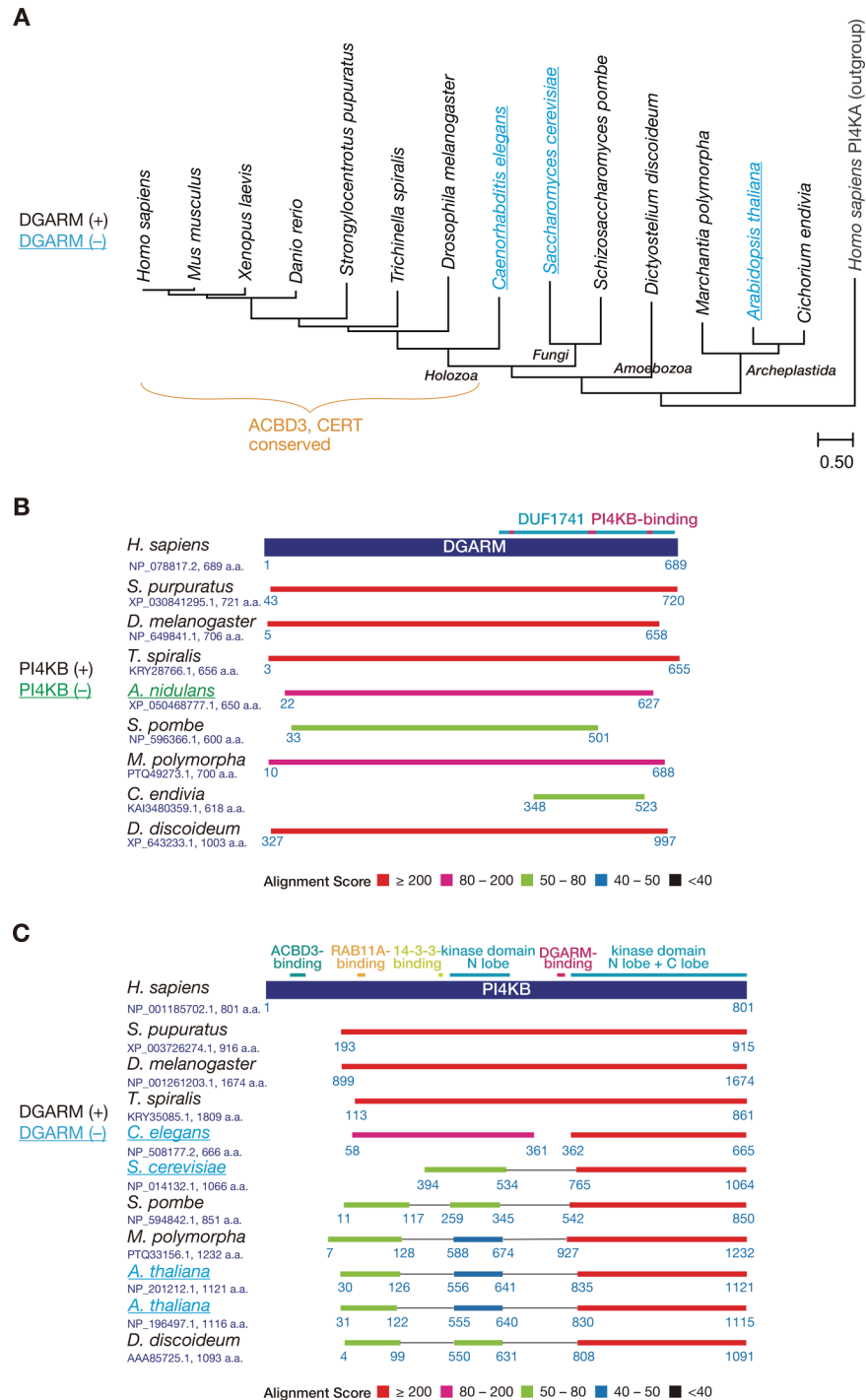


Figure 3. Evolutionary conservation of the DGARM–PI4KB axis. (A) The molecular phylogenetic tree of PI4KB is shown. Primary sequences of PI4KB orthologs (and human PI4KA as an outgroup) registered in UniProt were aligned by MUSCLE program and subjected to generate the phylogenetic tree by maximum likelihood method using MEGA software (ver. 11). Scale bar of the branch length represents the number of changes per sites. The analysis of DGARM phylogeny was extended from Chan et al. (2019), while PSI-BLAST was used instead. It should also be noted that, while ortholog search suggested that some plant species do conserve DGARM ortholog, most of the plant species have lost it. (B and C) Distributions of the BLAST hit sequences on human DGARM (B) and PI4KB (C) in non-vertebrates. The accession number and length (a.a.) of protein sequences are shown below the species name. The numbering represents the amino acid number. Domains of the query proteins are shown above as colored lines. Note that *A. thaliana* genome encodes two copies of *PI4KB* homolog. For ortholog search, PSI-BLAST was executed against the non-redundant protein sequences database for selected organisms with the profile-inclusion threshold of e-value set to 0.005. Species that lack DGARM or PI4KB are underlined and colored in cyan or green, respectively.

Fromme, 2021). On the other hand, the Frq1 ortholog in another model yeast *Schizosaccharomyces pombe*, which has DGARM, is dispensable for Golgi localization of Pik1 (Willet et al., 2023). However, whether DGARM interacts with Pik1 in *S. pombe* (and other fungal, plant, and ameba species) needs to be analyzed, as the DGARM-binding region is not conserved in Pik1 of these species. It should also be noted that some of the filamentous fungi (e.g., *Aspergillus nidulans*) are unlikely to have a Pik1 ortholog, whereas they have a DGARM. Other PtdIns 4-kinase(s) could be responsible for PtdIns(4)P production at the late Golgi although it remains unclear whether DGARM orthologs contribute to the generation of Golgi PtdIns(4)P pools in these species.

One of the pivotal roles of PtdIns(4)P in the distal Golgi membrane is to determine the contact sites with the ER, as mentioned above. The ER-distal-Golgi contacts provide the cell with a new pathway for the direct delivery of lipids from the ER to the distal Golgi regions, bypassing the proximal regions. The pathway that explicitly demands to bypass the proximal Golgi is ceramide delivery for *de novo* synthesis of phosphosphingolipids in vertebrates and insects (Figure 2A). Sphingomyelin synthase and insect-specific CPE synthase localize at the distal Golgi (Halter et al., 2007; Vacaru et al., 2013), whereas glucosylceramide synthase, which competes for the precursor ceramide, occurs predominantly at the proximal Golgi (Allan and Obradors, 1999; Hayashi et al., 2018; Ishibashi et al., 2018). Therefore, vertebrates and insects might require direct ceramide supply from the ER for robust phosphosphingolipid synthesis (Hanada et al., 2003; Rao et al., 2007). Notably, the evolutionary emergence of CERT is nearly coincident with that of SM and its close relatives including CPE (Hanada, 2014; Sato et al., 2019).

Perspectives

Local PtdIns(4)P production at the distal Golgi was suggested to be essential for growth and development since homozygous knockout of *DGARM* caused embryonic lethality in mice (Perez-Garcia et al., 2018). However, there remain unsolved questions about the regulatory mechanism of DGARM. (a) What factor(s) does DGARM recognize to focally localize at sites appropriate for lipid transport in the distal Golgi? (b) DGARM was suggested to cycle between the cytosol and Golgi membrane (Blomen et al., 2015; Chan et al., 2019), but what switches the localization of DGARM? Future studies on these issues would help us comprehend the details of the regulation system of the ER–distal Golgi contacts, which are presumed to be highly dynamic and short-lived (David et al., 2021; Subra et al., 2023). Evolutional analysis for the function of DGARM orthologs, especially in fungi and plants that do not possess CERT, would also broaden our understanding of the significance of constructing the transport pathway which bypasses the proximal Golgi.

A number of studies utilizing genome-wide knockout screens for various cellular events, including pathogen infection, have identified DGARM as a candidate (Blomen et al., 2015; Jeng et al., 2019; Yang et al., 2023, Cigler et al., 2023; Sasaki et al., 2023). Studies on the functional roles of DGARM in these cellular events would also attract interest in the pathologic and therapeutic aspects.

Abbreviations

CPE	ceramide phosphoethanolamine
ER	endoplasmic reticulum
ERGIC	ER–Golgi intermediate compartment
FFAT	two phenylalanines in an acidic tract
IPC	inositol phosphorylceramide
LTPs	lipid transfer proteins
ORD	OSBP-related domain
ORPs	OSBP-related proteins
PH	pleckstrin homology
PtdIns	phosphatidylinositol
PtdIns(4)P	phosphatidylinositol 4-monophosphate
START	steroidogenic acute regulatory protein-related lipid transfer
TGN	<i>trans</i> -Golgi network

Acknowledgement

We would like to deeply thank the collaborators for their invaluable contribution on the study of DGARM and CERT.

Declaration of Conflicting Interests


The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The authors disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work was supported by the Japan Agency for Medical Research and Development, Japan Society for the Promotion of Science (grant number JP22fk0108561, JP21H02630).

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References

- Allan D, Obradors MJM (1999). Enzyme distributions in subcellular fractions of BHK cells infected with semliki forest virus: Evidence for a major fraction of sphingomyelin synthase in the *trans*-Golgi network. *Biochim Biophys Acta - Mol Cell Res* 1450, 277–287. [https://doi.org/10.1016/S0167-4889\(99\)00057-9](https://doi.org/10.1016/S0167-4889(99)00057-9)
- Anwar MU, Sergeeva OA, Abrami L, Mesquita FS, Lukonin I, Amen T, Chuat A, Capolupo L, Liberali P, D'Angelo G, van der Goot FG (2022). ER-Golgi-localized proteins TMED2 and TMED10 control the formation of plasma membrane lipid nano-domains. *Dev Cell* 57, 2334–2346. <https://doi.org/10.1016/j.devcel.2022.09.004>

- Balla A, Tuymetova G, Barshishat M, Geiszt M, Balla T (2002). Characterization of type II phosphatidylinositol 4-kinase isoforms reveals association of the enzymes with endosomal vesicular compartments. *J Biol Chem* 277, 20041–20050. <https://doi.org/10.1074/jbc.M111807200>
- Balla T, Kim YJ, Alvarez-Prats A, Pemberton J (2019). Lipid dynamics at contact sites between the endoplasmic reticulum and other organelles. *Annu Rev Cell Dev Biol* 35, 85–109. <https://doi.org/10.1146/annurev-cellbio-100818-125251>
- Blomen VA, Majek P, Jae LT, Bigenzahn JW, Nieuwenhuis J, Staring J, Sacco R, van Diemen FR, Olk N, Stukalov A, et al. (2015). Gene essentiality and synthetic lethality in haploid human cells. *Science* 350, 1092–1096. <https://doi.org/10.1126/science.aac7557>
- Chan CJ, Le R, Burns K, Ahmed K, Coyaud E, Laurent EMN, Raught B, Melançon P (2019). BioID performed on Golgi enriched fractions identify C10orf76 as a GBF1 binding protein essential for Golgi maintenance and secretion. *Mol Cell Proteomics* 18, 2285–2297. <https://doi.org/10.1074/mcp.RA119.001645>
- Daboussi L, Costaguta G, Ghukasyan R, Payne GS (2017). Conserved role for Gga proteins in phosphatidylinositol 4-kinase localization to the *trans*-Golgi network. *Proc Natl Acad Sci* 114, 3433–3438. <https://doi.org/10.1073/pnas.1615163114>
- D'Angelo G, Polishchuk E, Di Tullio G, Santoro M, Di Campli A, Godi A, West G, Bielawski J, Chuang CC, Van Der Spoel AC, et al. (2007). Glycosphingolipid synthesis requires FAPP2 transfer of glucosylceramide. *Nature* 449, 62–67. <https://doi.org/10.1038/nature06097>
- D'Angelo G, Vicinanza M, Campli AD, De Matteis MA (2008). The multiple roles of PtdIns(4)P - not just the precursor of PtdIns(4,5)P₂. *J Cell Sci* 121, 1955–1963. <https://doi.org/10.1242/jcs.023630>
- David Y, Castro IG, Schuldiner M (2021). The fast and the furious: Golgi contact sites. *Contact* 4, 1–15. <https://doi.org/10.1177/25152564211034424>
- Del Bel LM, Brill JA (2018). Sac1, a lipid phosphatase at the interface of vesicular and nonvesicular transport. *Traffic* 19, 301–318. <https://doi.org/10.1111/tra.12554>
- de Saint-Jean M, Delfosse V, Douguet D, Chicanne G, Payrastra B, Bourguet W, Antonny B, Drin G (2011). Osh4p exchanges sterols for phosphatidylinositol 4-phosphate between lipid bilayers. *J Cell Biol* 195, 965–978. <https://doi.org/10.1083/jcb.201104062>
- Dickson EJ, Hille B (2019). Understanding phosphoinositides: Rare, dynamic, and essential membrane phospholipids. *Biochem J* 476, 1–23. <https://doi.org/10.1042/BCJ20180022>
- Doray B, Ghosh P, Griffith J, Geuze HJ, Kornfeld S (2002). Cooperation of GGAs and AP-1 in packaging MPRs at the *trans*-Golgi network. *Science* 297, 1700–1703. <https://doi.org/10.1126/science.1075327>
- Fang R, Jiang Q, Jia X, Jiang Z (2023). ARMH3-mediated Recruitment of PI4KB directs Golgi-to-endosome trafficking and activation of the antiviral effector STING. *Immunity* 56, 500–515. <https://doi.org/10.1016/j.immuni.2023.02.004>
- Goto A, Mizuike A, Hanada K (2020). Sphingolipid metabolism at the ER-Golgi contact zone and its impact on membrane trafficking. *Contact* 3, 1–13. <https://doi.org/10.1177/2515256420959514>
- Halter D, Neumann S, van Dijk SM, Wolthoorn J, de Mazière AM, Vieira OV, Mattjus P, Klumperman J, van Meer G, Sprong H (2007). Pre- and post-Golgi translocation of glucosylceramide in glycosphingolipid synthesis. *J Cell Biol* 179, 101–115. <https://doi.org/10.1083/jcb.200704091>
- Hanada K (2014). Co-evolution of sphingomyelin and the ceramide transport protein CERT. *Biochim Biophys Acta - Mol Cell Biol Lipids* 1841, 704–719. <https://doi.org/10.1016/j.bbalip.2013.06.006>
- Hanada K (2018). Lipid transfer proteins rectify inter-organelle flux and accurately deliver lipids at membrane contact sites. *J Lipid Res* 59, 1341–1366. <https://doi.org/10.1194/jlr.R085324>
- Hanada K, Kumagai K, Yasuda S, Miura Y, Kawano M, Fukasawa M, Nishijima M (2003). Molecular machinery for non-vesicular trafficking of ceramide. *Nature* 426, 803–809. <https://doi.org/10.1038/nature02188>
- Hayashi Y, Nemoto-Sasaki Y, Matsumoto N, Hama K, Tanikawa T, Oka S, Saeki T, Kumasaka T, Koizumi T, Arai S, et al. (2018). Complex formation of sphingomyelin synthase 1 with glucosylceramide synthase increases sphingomyelin and decreases glucosylceramide levels. *J Biol Chem* 293, 17505–17522. <https://doi.org/10.1074/jbc.RA118.002048>
- Highland CM, Fromme JC (2021). Arf1 directly recruits the Pik1-Frq1 PI4K complex to regulate the final stages of Golgi maturation. *Mol Biol Cell* 32, 1064–1080. <https://doi.org/10.1091/mbc.E21-02-0069>
- Huitema K, Van Den Dikkenberg J, Brouwers JFHM, Holthuis JCM (2004). Identification of a family of animal sphingomyelin synthases. *EMBO J* 23, 33–44. <https://doi.org/10.1038/sj.emboj.7600034>
- Ishibashi Y, Ito M, Hirabayashi Y (2018). Regulation of glucosylceramide synthesis by Golgi-localized phosphoinositide. *Biochem Biophys Res Commun* 499, 1011–1018. <https://doi.org/10.1016/j.bbrc.2018.04.039>
- Jeng EE, Bhadkamkar V, Ibe NU, Gause H, Jiang L, Chan J, Jian R, Jimenez-Morales D, Stevenson E, Krogan NJ, et al. (2019). Systematic identification of host cell regulators of *Legionella pneumophila* pathogenesis using a genome-wide CRISPR screen. *Cell Host Microbe* 26, 551–563. <https://doi.org/10.1016/j.chom.2019.08.017>
- Jumper J, Evans R, Pritzel A, Green T, Figurnov M, Ronneberger O, Tunyasuvunakool K, Bates R, Žídek A, Potapenko A, et al. (2021). Highly accurate protein structure prediction with AlphaFold. *Nature* 596, 583–589. <https://doi.org/10.1038/s41586-021-03819-2>
- Kajiwara K, Ikeda A, Aguilera-Romero A, Castillon GA, Kagiwada S, Hanada K, Riezman H, Muniz M, Funato K (2014). Osh proteins regulate COPII-mediated vesicular transport of ceramide from the endoplasmic reticulum in budding yeast. *J Cell Sci* 127, 376–387. <https://doi.org/10.1242/jcs.132001>
- Limar S, Körner C, Martínez-Montañés F, Stancheva VG, Wolf VN, Walter S, Miller EA, Ejsing CS, Galassi VV, Fröhlich F (2023). Yeast Svf1 binds ceramides and contributes to sphingolipid metabolism at the ER cis-Golgi interface. *J Cell Biol* 222, e202109162. <https://doi.org/10.1083/jcb.202109162>
- Liu LK, Choudhary V, Toulmay A, Prinz WA (2017). An inducible ER-Golgi tether facilitates ceramide transport to alleviate lipotoxicity. *J Cell Biol* 216, 131–147. <https://doi.org/10.1083/jcb.201606059>
- Lu D, Sun H, Wang H, Barylko B, Fukata Y, Fukata M, Albanesi JP, Yin HL (2012). Phosphatidylinositol 4-Kinase II α Is Palmitoylated by Golgi-localized Palmitoyltransferases in

- cholesterol-dependent manner. *J Biol Chem* 287, 21856–21865. <https://doi.org/10.1074/jbc.M112.348094>
- McPhail JA, Burke JE (2023). Molecular mechanisms of PI4K regulation and their involvement in viral replication. *Traffic* 24, 131–145. <https://doi.org/10.1111/tra.12841>
- McPhail JA, Lyoo H, Pemberton JG, Hoffmann RM, Elst W, Strating JR, Jenkins ML, Stariha JT, Powell CJ, Boulanger MJ, et al. (2020). Characterization of the c10orf76-PI4KB complex and its necessity for Golgi PI4P levels and enterovirus replication. *EMBO Rep* 21, e48441. <https://doi.org/10.15252/embr.201948441>
- Mesmin B, Bigay J, Moser von Filseck J, Lacas-Gervais S, Drin G, Antony B (2013). A four-step cycle driven by PI(4)P hydrolysis directs sterol/PI(4)P exchange by the ER-Golgi tether OSBP. *Cell* 155, 830–843. <https://doi.org/10.1016/j.cell.2013.09.056>
- Mizuike A, Sakai S, Katoh K, Yamaji T, Hanada K (2023). The C10orf76–PI4KB axis orchestrates CERT-mediated ceramide trafficking to the distal Golgi. *J Cell Biol* 222, e202111069. <https://doi.org/10.1083/jcb.202111069>
- Mogelsvang S, Marsh BJ, Ladinsky MS, Howell KE (2004). Predicting function from structure: 3D structure studies of the mammalian Golgi complex. *Traffic* 5, 338–345. <https://doi.org/10.1111/j.1398-9219.2004.00186.x>
- Moreau P, Cassagne C (1994). Phospholipid trafficking and membrane biogenesis. *Biochim Biophys Acta - Rev Biomembr* 1197, 257–290. [https://doi.org/10.1016/0304-4157\(94\)90010-8](https://doi.org/10.1016/0304-4157(94)90010-8)
- Motani K, Saito-Tarashima N, Nishino K, Yamauchi S, Minakawa N, Kosako H (2022). The Golgi-resident protein ACBD3 concentrates STING at ER-Golgi contact sites to drive export from the ER. *Cell Rep* 41, 111868. <https://doi.org/10.1016/j.celrep.2022.111868>
- Mukai K, Konno H, Akiba T, Uemura T, Waguri S, Kobayashi T, Barber GN, Arai H, Taguchi T (2016). Activation of STING requires palmitoylation at the Golgi. *Nat Commun* 7, 11932. <https://doi.org/10.1038/ncomms11932>
- Nishitoh H (2019). Paradigm shift from ‘compartment’ to ‘zone’ in the understanding of organelles. *J Biochem* 165, 97–99. <https://doi.org/10.1093/jb/mvy107>
- Panevska A, Skočaj M, Križaj I, Maček P, Sepčić K (2019). Ceramide phosphoethanolamine, an enigmatic cellular membrane sphingolipid. *Biochim Biophys Acta - Biomembr* 1861, 1284–1292. <https://doi.org/10.1016/j.bbamem.2019.05.001>
- Pantazopoulou A, Glick BS (2019). A kinetic view of membrane traffic pathways can transcend the classical view of Golgi compartments. *Front Cell Dev Biol* 7, 153. <https://doi.org/10.3389/fcell.2019.00153>
- Perez-Garcia V, Fineberg E, Wilson R, Murray A, Mazzeo CI, Tudor C, Sienerth A, White JK, Tuck E, Ryder EJ, et al. (2018). Placentation defects are highly prevalent in embryonic lethal mouse mutants. *Nature* 555, 463–468. <https://doi.org/10.1038/nature26002>
- Perry RJ, Ridgway ND (2006). Oxysterol-binding protein and vesicle-associated membrane protein-associated protein are required for sterol-dependent activation of the ceramide transport protein. *Mol Biol Cell* 17, 2604–2616. <https://doi.org/10.1091/mbc.e06-01-0060>
- Posor Y, Jang W, Haucke V (2022). Phosphoinositides as membrane organizers. *Nat Rev Mol Cell Biol* 23, 797–816. <https://doi.org/10.1038/s41580-022-00490-x>
- Rao RP, Yuan C, Allegood JC, Rawat SS, Edwards MB, Wang X, Merrill AH, Acharya U, Acharya JK (2007). Ceramide transfer protein function is essential for normal oxidative stress response and lifespan. *Proc Natl Acad Sci* 104, 11364–11369. <https://doi.org/10.1073/pnas.0705049104>
- Ritchie C, Carozza JA, Li L (2022). Biochemistry, cell biology, and pathophysiology of the innate immune cGAS-cGAMP-STING pathway. *Annu Rev Biochem* 91, 599–628. <https://doi.org/10.1146/annurev-biochem-040320-101629>
- Sasaki J, Ishikawa K, Arita M, Taniguchi K (2012). ACBD3-mediated Recruitment of PI4KB to picornavirus RNA replication sites. *EMBO J* 31, 754–766. <https://doi.org/10.1038/emboj.2011.429>
- Sasaki K, Yoshida H (2019). Organelle zones. *Cell Struct Funct* 44, 85–94. <https://doi.org/10.1247/csf.19010>
- Sato M, Arita M, Kawashima T (2019). Uncovering ecdysozoa-specific sphingomyelin synthase by phylogenetic analysis of metazoan sequences. *Zool Sci* 36, 316–321. <https://doi.org/10.2108/zs180168>
- Strahl T, Hama H, DeWald DB, Thorner J (2005). Yeast phosphatidylinositol 4-kinase, Pik1, has essential roles at the Golgi and in the nucleus. *J Cell Biol* 171, 967–979. <https://doi.org/10.1083/jcb.200504104>
- Subra M, Dezi M, Bigay J, Lacas-Gervais S, Cicco AD, Araújo ARD, Abélanet S, Fleuriot L, Debayle D, Gautier R, et al. (2023). VAP-A intrinsically disordered regions enable versatile tethering at membrane contact sites. *Dev Cell* 58, 121–138. <https://doi.org/10.1016/j.devcel.2022.12.010>
- Suda Y, Nakano A (2012). The Yeast Golgi apparatus. *Traffic* 13, 505–510. <https://doi.org/10.1111/j.1600-0854.2011.01316.x>
- Tewari R, Bailes E, Bunting KA, Coates JC (2010). Armadillo-repeat protein functions: Questions for little creatures. *Trends Cell Biol* 20, 470–481. <https://doi.org/10.1016/j.tcb.2010.05.003>
- Tie HC, Ludwig A, Sandin S, Lu L (2018). The spatial separation of processing and transport functions to the interior and periphery of the Golgi stack. *Elife* 7, 1–26. <https://doi.org/10.7554/eLife.41301>
- Vacaru AM, Van Den Dikkenberg J, Ternes P, Holthuis JCM (2013). Ceramide phosphoethanolamine biosynthesis in *Drosophila* is mediated by a unique ethanolamine phosphotransferase in the Golgi lumen. *J Biol Chem* 288, 11520–11530. <https://doi.org/10.1074/jbc.M113.460972>
- Varadi M, Anyango S, Deshpande M, Nair S, Natassia C, Yordanova G, Yuan D, Stroe O, Wood G, Laydon A, et al. (2022). AlphaFold protein structure database: Massively expanding the structural coverage of protein-sequence space with high-accuracy models. *Nucleic Acids Res* 50, D439–D444. <https://doi.org/10.1093/nar/gkab1061>
- Venditti R, Rega LR, Masone MC, Santoro M, Polishchuk E, Sarnataro D, Paladino S, D’Auria S, Varriale A, Olkkonen VM, et al. (2019). Molecular determinants of ER-Golgi contacts identified through a new FRET-FLIM system. *J Cell Biol* 218, 1055–1065. <https://doi.org/10.1083/jcb.201812020>
- Willet AH, Turner LA, Park JS, Ren L, Snider CE, Gould KL (2023). Characterization of Pik1 function in fission yeast reveals its conserved role in lipid synthesis and not cytokinesis. *J Cell Sci* 136, e00146. <https://doi.org/10.1242/jcs.261415>
- Wong LH, Gatta AT, Levine TP (2019). Lipid transfer proteins: The lipid commute via shuttles, bridges and tubes. *Nat Rev Mol Cell Biol* 20, 85–101. <https://doi.org/10.1038/s41580-018-0071-5>

- Yang J, Guo F, Chin HS, Bin Chen G, Ang CH, Lin Q, Hong W, Fu NY (2023). Sequential genome-wide CRISPR-Cas9 screens identify genes regulating cell-surface expression of tetraspanins. *Cell Rep* 42, 112065. <https://doi.org/10.1016/j.celrep.2023.112065>
- Zhang C, Shang G, Gui X, Zhang X, Chen Bai X, Chen ZJ (2019). Structural basis of STING binding with and phosphorylation by TBK1. *Nature* 567, 394–398. <https://doi.org/10.1038/s41586-019-1000-2>