© 2016. Published by The Company of Biologists Ltd | Journal of Cell Science (2016) 129, 4329-4339 doi:10.1242/jcs.196287

COMMENTARY

Mechanisms and functions of lysosome positioning

Jing Pu, Carlos M. Guardia, Tal Keren-Kaplan and Juan S. Bonifacino*

ABSTRACT

Lysosomes have been classically considered terminal degradative organelles, but in recent years they have been found to participate in many other cellular processes, including killing of intracellular pathogens, antigen presentation, plasma membrane repair, cell adhesion and migration, tumor invasion and metastasis, apoptotic cell death, metabolic signaling and gene regulation. In addition, lysosome dysfunction has been shown to underlie not only rare lysosome storage disorders but also more common diseases, such as cancer and neurodegeneration. The involvement of lysosomes in most of these processes is now known to depend on the ability of lysosomes to move throughout the cytoplasm. Here, we review recent findings on the mechanisms that mediate the motility and positioning of lysosomes, and the importance of lysosome dynamics for cell physiology and pathology.

KEY WORDS: Lysosome, Late endosome, Organelle positioning, Organelle motility, Kinesin, Dynein

Introduction

Lysosomes are membrane-enclosed cytoplasmic organelles responsible for the degradation of a variety of biomacromolecules in the endomembrane system of animal cells (Braulke and Bonifacino, 2009; Saftig and Klumperman, 2009). The degradative function of lysosomes is carried out by over 60 luminal hydrolases with specificity for different substrates. Most of these hydrolases have an acidic optimum pH similar to that of the lysosome lumen (4.5–5.0). The limiting membrane of lysosomes comprises more than 200 integral membrane proteins, including a proton-importing V-type ATPase that maintains the acidic pH of the lumen, ion transporters and channels that regulate the luminal ion composition, multiple other transporters that export the products of degradation to the cytosol, a set of highly-glycosylated lysosome-associated membrane proteins (LAMPs) that protect the membrane from degradation by the lysosomal hydrolases, and tethering factors and SNARE proteins that promote contact and fusion, respectively, of lysosomes with other cytoplasmic organelles. Finally, the cytosolic face of the lysosomal membrane serves as a platform for numerous proteins and protein complexes that mediate nutrient and stress signaling, as well as interactions with the cytoskeleton.

Lysosomes can degrade materials delivered to them by way of biosynthetic transport, endocytosis, phagocytosis and autophagy. Because of their prominent role in intracellular degradation, lysosomes have earned a reputation as the ‘garbage disposal system’ of the cell. In recent years, however, this perception of lysosomes as mere degradative organelles has started to change with the realization that they participate in many other cellular processes, including killing of intracellular pathogens, antigen presentation, plasma membrane repair, exosome release, cell adhesion and migration, tumor invasion and metastasis, apoptosis, metabolic signaling and gene regulation (Ballabio, 2016; Mukhopadhyay, 2016) (Fig. 1). In addition, lysosomal defects have been shown to underlie not only rare lysosomal storage disorders (LSDs), but also common neurodegenerative disorders such as Alzheimer’s, Parkinson’s and Huntington’s diseases, as well as amyotrophic lateral sclerosis (ALS) (Parenti et al., 2015; Fraldi et al., 2016). These findings have rekindled interest in lysosomes as crucial effectors of multiple cellular functions.

In this Commentary, we discuss an emerging property of lysosomes that influences many of their functions: their positioning and motility within the cytoplasm. Because late endosomes and lysosomes share many properties, in most cases we refer to both organelles as ‘lysosomes’, except when a distinction is necessary.

Lysosome positioning and motility

Lysosomes are broadly distributed throughout the cytoplasm (Fig. 2). In non-polarized cells, they are most concentrated in a central region surrounding the microtubule-organizing center (MTOC) (the ‘perinuclear cloud’) (Jongsma et al., 2016), although there are also many peripheral lysosomes, some reaching as far as the plasma membrane and cell protrusions. In polarized cells such as neurons, lysosomes are also found in all cytoplasmic domains including the soma, axon and dendrites. Although some lysosomes are relatively static, others move bidirectionally along microtubule tracks between the center and the periphery of the cell (Matteoni and Kreis, 1987). Lysosome movement towards the microtubule plus-ends (anterograde) and minus-ends (retrograde) is mediated by kinesin (Hollenbeck and Swanson, 1990) and dynein motors (Harada et al., 1998), respectively. In general, lysosome movement is not continuous but ‘stop-and-go’ (Jordens et al., 2001), suggesting that it is subject to regulation. Moreover, the distribution and motility of lysosomes change under certain conditions. For example, cytosol acidification causes dispersal of the perinuclear lysosome population, whereas subsequent alkalinization returns them to their central location (Heuser, 1989; Parton et al., 1991). Other perturbations, such as aggresome formation (Zaarur et al., 2014), starvation (Korolchuk et al., 2011), drug-induced apoptosis (Yu et al., 2016), expression of pathogenic mutant forms of huntingtin (Erie et al., 2015) or leucine-rich repeat kinase 2 (LRRK2) (Dodson et al., 2012) and LSDs (Uusi-Rauva et al., 2012; Li et al., 2016b), result in perinuclear clustering of lysosomes. During movement, lysosomes sometimes tubulate in a process that might involve attachment to both kinesins and dynein pulling in opposite directions (Mrakovic et al., 2012; Li et al., 2016b). Lysosome dispersal and tubulation can also be induced by specific stimuli. For example, treatment of macrophages or dendritic cells with bacterial lipopolysaccharide (a surrogate for bacterial infection) causes massive lysosome tubulation, a process that contributes to phagosome maturation and antigen presentation

Cell Biology and Neurobiology Branch, Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD 20892, USA.

*Author for correspondence (bonifacinoj@helix.nih.gov)

J.S.B., 0000-0002-5673-6370

Journal of Cell Science (2016) 129, 4329-4339 doi:10.1242/jcs.196287

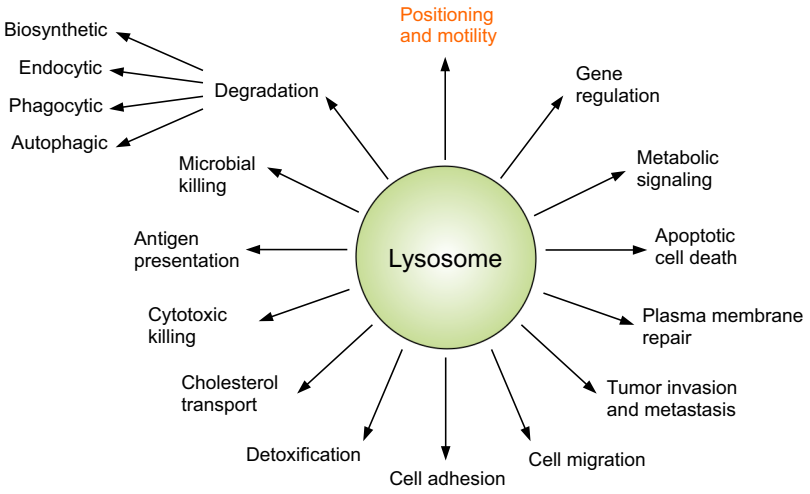

Fig. 1. Multiple functions of lysosomes are influenced by their positioning and motility. In addition to degradation, lysosomes participate in other cellular functions. Many of these functions, including endocytic, phagocytic and autophagic degradation, antigen presentation, killing of target cells by cytotoxic T-cells and NK cells, metabolic signaling, cell adhesion and migration, tumor invasion and metastasis, as well as plasma membrane repair, depend on the ability of lysosomes to move throughout the cytoplasm.

(Mrakovic et al., 2012; Saric et al., 2016). Similarly, dendritic cell maturation induces lysosome tubulation for delivery of major histocompatibility complex (MHC) class II (MHC-II) molecules to the cell surface (Chow et al., 2002; Vyas et al., 2007). Lysosome movement is also modulated by contacts with other organelles, such as the endoplasmic reticulum (ER) (Raiborg et al., 2015; Jongsma et al., 2016), the trans-Golgi network (TGN) (Wang and Hong, 2002) and peroxisomes (Chu et al., 2015). Lysosome movement allows lysosomes to survey the entire cytoplasmic space, both constitutively and in a regulated manner, in order to perform their multiple functions.

Anterograde transport

Kinesins are microtubule motors involved in the movement of multiple cytoplasmic organelles, including lysosomes (Hirokawa and Noda, 2008). Mammalian genomes encode about 45 kinesin superfamily (KIF) proteins, and many more variants are generated by alternative splicing of the corresponding mRNAs. All KIFs comprise a globular motor domain that attaches to microtubules, and a tail domain that interacts with specific adaptors or cargos. ATP hydrolysis by the motor domain provides the driving force for translocation of kinesins and associated cargos from the minus end to the plus end of microtubules (i.e. anterograde transport), with the exception of kinesin-14 members, which move in the plus-to-minus-end direction (Hirokawa and Noda, 2008). In non-polarized cells, microtubule plus-ends are generally found in the peripheral cytoplasm and in neurons they point towards axon terminals, so, in these cells, kinesins mediate transport from the cell center towards the periphery (known as centrifugal transport). In some polarized cells, however, microtubules can point in other directions, as is the case for neuronal dendrites, which have microtubules with mixed orientations (Baas et al., 1988; Yau et al., 2016). In these cases, kinesins can potentially mediate both centrifugal and centripetal transport.

Remarkably, lysosome movement has been shown to depend on not one but multiple kinesins, including kinesin-1 (KIF5A, KIF5B and KIF5C) (Nakata and Hirokawa, 1995; Tanaka et al., 1998; Rosa-Ferreira and Munro, 2011), kinesin-2 (KIF3) (Brown et al., 2005; Loubéry et al., 2008) and kinesin-3 (KIF1A and KIF1B) (Matsushita et al., 2004; Korolchuk et al., 2011; Bentley et al., 2015), as well as the kinesin-13 (KIF2) family members (Santama et al., 1998; Korolchuk et al., 2011) (Fig. 3). At present it is unclear why so many kinesins have evolved to move the same organelle. Possible explanations are (1) functional redundancy, (2) cell-type-specific expression (e.g. non-neuronal versus neuronal cells), (3) involvement in different lysosomal functions (e.g. autophagy versus exocytosis), (4) differential regulation, and (5) transport along different microtubule tracks. With regard to this latter possibility, several kinesins exhibit preferential association with microtubule tracks that are characterized by specific post-translational

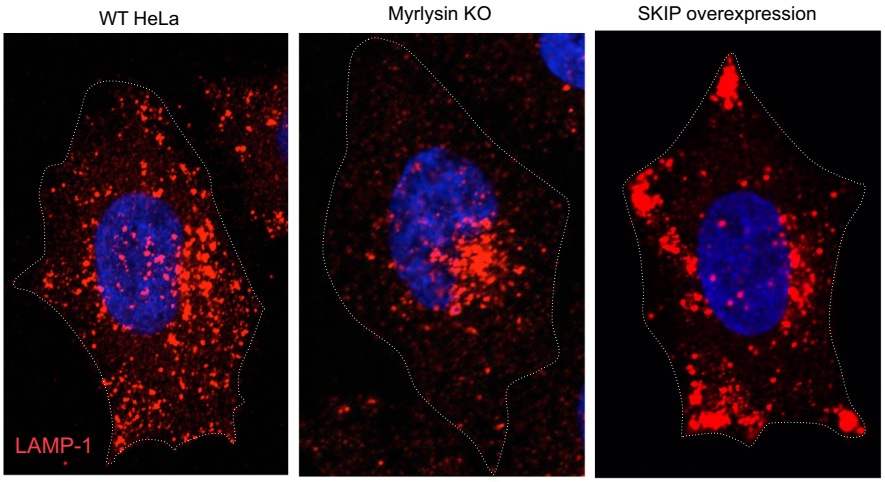

Fig. 2. Distribution of lysosomes in HeLa cells. Wild-type (WT), myrlysin-knockout (KO), and SKIP-overexpressing HeLa cells were immunostained for the lysosomal membrane protein LAMP-1. In WT cells, lysosomes are scattered throughout the cytoplasm, although they have a greater concentration in the juxtanuclear area of the cells. In cells lacking the myrlysin subunit of BORC, lysosomes are more clustered in the juxtanuclear area and depleted from the periphery. In SKIP-overexpressing cells, lysosomes accumulate in the periphery. See Fig. 4 for a schematic representation of the roles of BORC and SKIP in lysosome positioning.

Journal of Cell Science (2016) 129, 4329-4339 doi:10.1242/jcs.196287

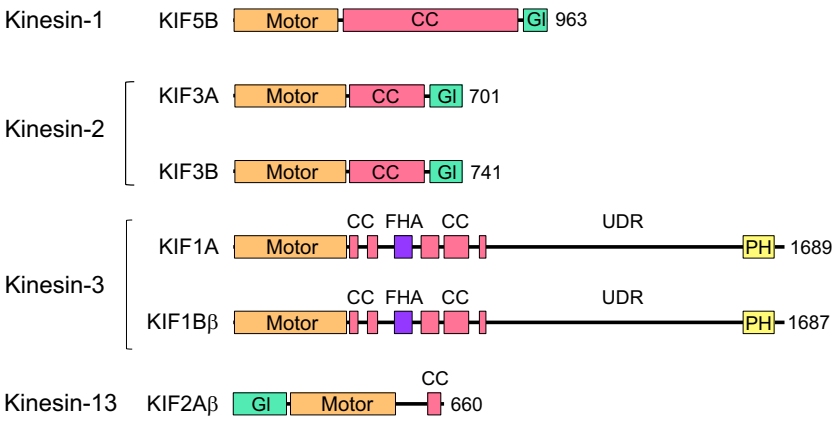

Fig. 3. Kinesins implicated in lysosome movement. Family names, domains and amino acid numbers are indicated. CC, coiled coil; GI, globular; FHA, forkhead-associated; UDR, undefined region; PH, pleckstrin-homology.

modifications (PTMs) of tubulin, microtubule-associated proteins (MAPs) or kinesin-binding proteins (KBPs) (Marx et al., 2005). For example, kinesin-1 motors move faster on microtubule tracks enriched in acetylated and GTP-bound tubulin, but slower motors that make longer processive runs have been observed on detyrosinated microtubules (Reed et al., 2006; Dunn et al., 2008; Hammond et al., 2008; Konishi and Setou, 2009; Nakata et al., 2011). Kinesin-2 (KIF17) and -3 (KIF1A) family members do not exhibit preferences for acetylated or detyrosinated microtubules (Cai et al., 2009), but KIF1A-dependent transport is influenced by tubulin polyglutamylation (Ikegami et al., 2007). MAPs often inhibit kinesin-dependent transport by acting as obstacles to movement. A prime example of this behavior is mediated by the protein tau, which blocks kinesin-1-dependent motility (Dehmelt and Halpain, 2005; Al-Bassam et al., 2007). Other MAPs (e.g. ensconsin and DCLK1), however, promote recruitment and activation of kinesins on specific populations of microtubules (Sung et al., 2008; Lipka et al., 2016). Finally, KBPs can modulate kinesin activity, as shown for the inactivation of the motor domains of the kinesin-3 KIF1A and kinesin-8 KIF18A by the protein KBP (also known as KIF1BP and KIAA1279) (Kevenaar et al., 2016). It remains to be determined how all of these factors influence the ability of different kinesins to mediate different patterns of lysosome movement.

By far the best-characterized kinesin involved in lysosome transport is kinesin-1 (Figs 3 and 4). This kinesin is a heterotetramer composed of two heavy chains (KIF5A, KIF5B or KIF5C) and two light chains (KLC1, KLC2, KLC3 or KLC4) (DeBoer et al., 2008). Kinesin-1 is recruited to lysosomes by a chain of interacting proteins, including the multisubunit BLOC-1-related complex (BORC), the

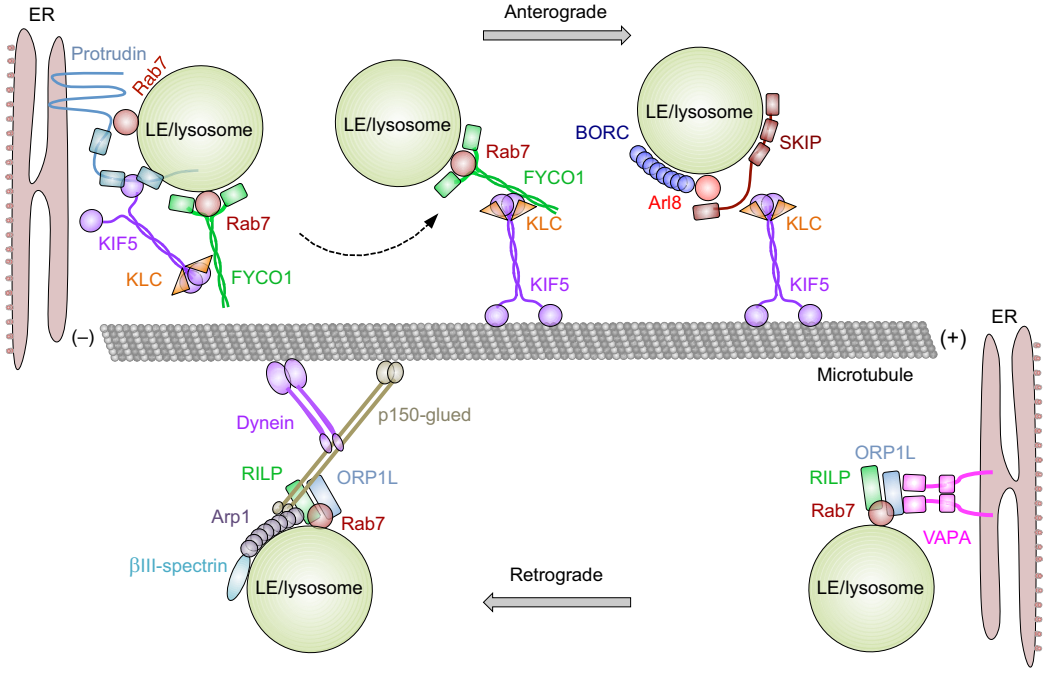

Fig. 4. Mechanisms of late endosome and lysosome transport along microtubules. Anterograde transport of late endosome (LE) and lysosome transport is mediated by an ensemble of BORC, Arl8, SKIP and kinesin-1 (a heterotetramer composed of two KLC and two KIF5 chains). An alternative mechanism of anterograde transport uses Rab7 and FYCO1 as adaptors to kinesin-1. FYCO1 is loaded onto late endosomes by the action of the ER-anchored protrudin. Other kinesins depicted in Fig. 3 have also been shown to drive anterograde transport of lysosomes, but their mechanisms of coupling are less well understood. Retrograde transport is mediated by Rab7, RILP, ORP1L and dynein–dynactin. The names of some of the dynactin subunits are indicated (p150-glued, Arp1). Under low-cholesterol concentrations, ORP1L interacts with the ER-anchored protein VAPA, leading to dynein dissociation and redistribution of lysosomes to the cell periphery.

Arf-like small GTPase Arl8 (which has two isoforms in mammals, Arl8a and Arl8b, hereafter generically referred to as Arl8), and the Arl8 effector SifA and kinesin-interacting protein (SKIP, also known as PLEKHM2) (Boucrot et al., 2005; Bagshaw et al., 2006; Hofmann and Munro, 2006; Rosa-Ferreira and Munro, 2011; Pu et al., 2015) (Fig. 4). BORC is an octameric complex comprising BLOS1, BLOS2 (also known as BLOC1S1 and BORCS1, and BLOC1S2 and BORCS2, respectively) and snapin (also known as BORCS3) subunits (all three of which are shared with the BLOC-1 complex involved in the biogenesis of lysosome-related organelles) (Falcon-Perez et al., 2002; Moriyama and Bonifacino, 2002; Starcevic and Dell'Angelica, 2004), plus the unique subunits KXD1 (also known as BORCS4), MEF2BNB (also known as BORCS8), myrlysin (also known as LOH12CR1 and BORCS5), lyspersin (also known as C17orf59 and BORCS6) and diaskedin (also known as C10orf32 and BORCS7) (Pu et al., 2015). BORC associates with the cytosolic face of lysosomes partly through the myristoylated N-terminus of myrlysin, and is subsequently required for recruitment of Arl8 from the cytosol (Pu et al., 2015). This function of BORC would be consistent with it being a guanine-nucleotide-exchange factor (GEF) (Rosa-Ferreira and Munro, 2011) for Arl8, but there is currently no biochemical evidence for this activity. SKIP in turn binds to Arl8 through an N-terminal RUN domain (Rosa-Ferreira and Munro, 2011). A WD motif in an unstructured region of SKIP then interacts with the tetratricopeptide repeat (TPR) domain of the KLC (Rosa-Ferreira and Munro, 2011), thus completing the linkage of lysosomes to KIF5 proteins. Various ways of negatively interfering with this chain of interactors inhibit lysosome movement towards the cell periphery, resulting in collapse of the entire lysosomal population to the cell center (Fig. 2). Conversely, overexpression of some components of this cascade, such as SKIP, causes accumulation of lysosomes at the cell periphery (Fig. 2). An alternative mechanism for coupling of late endosomes to kinesin-1 involves the ER-anchored protein protrudin, which binds simultaneously to the small GTPase Rab7 (which has two isoforms in mammals, Rab7a and Rab7b, hereafter generically referred to as Rab7) and phosphatidylinositol 3-phosphate [PtdIns(3)*P*] to bridge the ER and lysosomal membranes. Protrudin then transfers late endosomes to the Rab7 effector FYVE- and coiled-coil-domain-containing protein (FYCO1) and kinesin-1 for late endosome movement towards the cell periphery (Matsuzaki et al., 2011; Raiborg et al., 2016) (Fig. 4).

A recent study has shown that the recruitment of the kinesin-3 proteins KIF1A and KIF1Bβ is also dependent on BORC and Arl8 (Guardia et al., 2016), as well as several parts of the C-terminal tail domain (Bentley et al., 2015; Guardia et al., 2016). Considerably less is known about the mechanisms that couple other kinesins to lysosomes. For the kinesin-2 KIF3A, the accessory protein KAP3 has been implicated in lysosome movement (Brown et al., 2005), but other potential regulators and adaptors remain to be identified. It would be of particular interest to investigate whether the Arl8- and Rab7-regulated pathways also operate for other kinesins involved in lysosome movement.

Retrograde transport

There are two types of dynein, an axonemal dynein that functions in cilia and flagella, and a cytoplasmic dynein that functions in the rest of the cell (Ishikawa, 2012). Cytoplasmic dynein (referred to as dynein for simplicity) moves organelles, including lysosomes, from the plus end to the minus end of microtubules (i.e. retrograde transport). In non-polarized cells, this results in organelle movement from the periphery to the cell center (i.e. centripetal) (Paschal and Vallee, 1987). In neurons, dynein also mediates centripetal transport in the axon, and potentially both centripetal and centrifugal transport in the dendrites because of the mixed orientation of dendritic microtubules. Dynein is a ∼1.4-MDa multimeric complex comprising two heavy chains, two intermediate chains, two light intermediate chains and several light chains. Dynein interacts with dynactin, another multimeric complex of ∼1.0 MDa composed of more than 20 subunits corresponding to 11 different proteins (Ishikawa, 2012). Dynein associates with lysosomes in a dynactin-dependent manner (Lin and Collins, 1992; Burkhardt et al., 1997) (Fig. 4).

The recruitment of dynein–dynactin to late endosomes and lysosomes is mainly dependent on Rab7. The nucleotide cycle of this Rab is regulated by the GTPase-activating proteins (GAPs) TBC1D15 (Zhang et al., 2005) and TBC1D2 (also known as Armus) (Frasa et al., 2010), and the GEF complex Mon1–Ccz1 (Nordmann et al., 2010). The homotypic fusion and protein sorting (HOPS) complex functions as a Rab7 GEF in yeast (Wurmser et al., 2000), and overexpression of some of the HOPS subunits causes perinuclear clustering of lysosomes in mammalian cells (Wurmser et al., 2000; Poupon et al., 2003). However, HOPS does not show GEF activity towards Rab7 in mammalian cells (Peralta et al., 2010). Thus, its effect on lysosome positioning must be mediated by a different mechanism, most likely by tethering to other organelles.

Several downstream effectors of Rab7 couple lysosomes to dynein–dynactin. One of these effectors is the Rab7-interacting lysosomal protein (RILP) (Cantalupo et al., 2001; Jordens et al., 2001; Progida et al., 2007), which links Rab7 to dynein–dynactin through the dynactin p150-glued (DCTN1) subunit (Johansson et al., 2007). An additional effector, the cholesterol sensor OSBP-related protein 1L (ORP1L, also known as OSBPL1A), forms a tripartite complex with Rab7 and RILP, promoting the association of membrane-bound βIII-spectrin with dynein–dynactin through the dynactin subunit actin-related protein 1 (Arp1, also known as ACTR1A) (Johansson et al., 2007) (Fig. 4). Interaction of ORP1L with both Rab7–RILP and βIII spectrin results in dynein motor activation. The ability of ORP1L to engage in these interactions is dependent on cholesterol levels. Under low-cholesterol conditions, the OSBP-related domain (ORD) of ORP1L binds the ER protein vesicle-associated membrane protein (VAMP)-associated ER protein A (VAPA), forcing dissociation of dynein–dynactin from lysosomes, and causing dispersal of these organelles to the cell periphery. In contrast, under high-cholesterol conditions, ORP1L does not interact with VAP, and the association of lysosomes with dynein–dynactin results in their movement towards the cell center (Rocha et al., 2009). Abnormal accumulation of cholesterol in late endosomes, which occurs in Niemann–Pick C disease, has also been reported to cause immobilization of these organelles in the perinuclear region (Ko et al., 2001; Lebrand et al., 2002), through interference with the Rab7 GTP-GDP cycle and kinesin function (Lebrand et al., 2002). In all of these cases, it is unclear how cholesterol is transferred from the lumen to the cytosolic leaflet of the lysosomal membrane to be sensed by ORP1L and Rab7.

Another Rab7 effector that causes lysosome clustering when overexpressed is Rabring7 (also known as RNF115) (Mizuno et al., 2003). This protein has a C-terminal RING finger motif with E3-ligase activity, suggesting an involvement of the ubiquitin pathway in lysosome positioning. The protein rapsyn also contains a RING finger and causes juxtanuclear lysosome clustering upon overexpression (Aittaleb et al., 2015). However, it is unknown whether Rabring7 and rapsyn function in conjunction with kinesins

or dynein, and what the precise role of ubiquitylation in this process might be.

Additional Rab proteins, including Rab9A, Rab34 and Rab36, have also been implicated in the regulation of lysosome positioning. RNA interference (RNAi)-mediated depletion of the lysosome-associated Rab9A causes juxtanuclear clustering of lysosomes (Ganley et al., 2004). Cholesterol accumulation in Niemann–Pick C disease also alters Rab9A activity and its association with late endosomes (Ganley and Pfeffer, 2006), suggesting that Rab9A might also be implicated in cholesterol-mediated regulation of late endosome positioning. Rab9A interacts with the PH domain of SKIP (Jackson et al., 2008) and of the Rab36 GAP RUTBC2 (also known as SGSM1) (Nottingham et al., 2012; Zhang et al., 2014), but it is unclear whether these interactions account for the role of Rab9A in lysosome positioning. Overexpression of Rab34 and Rab36 causes lysosome clustering (Wang and Hong, 2002; Chen and Yu, 2013). Both of these Rab proteins interact with RILP and thus mediate coupling to dynein, but they localize to the Golgi complex and/or TGN rather than lysosomes. This suggests that Rab34 and Rab36 promote the juxtanuclear localization of lysosomes through contacts with the Golgi or TGN.

In addition to Rab proteins, other proteins regulate coupling of lysosomes to dynein–dynactin. For example, the lysosomal Ca^{2+}-sensor ALG-2 (also known as PDCD6) promotes interaction of the lysosomal Ca^{2+} channel transient receptor potential mucolipin 1 (TRPMNL1 or MCOLN1) with dynein–dynactin to regulate centripetal transport of lysosomes in response to changes in Ca^{2+} levels (Li et al., 2016b). There are also lysosomal transmembrane proteins that regulate retrograde transport, such as LAMP-1 and LAMP-2, which promote coupling of lysosomes to dynein–dynactin (Huynh et al., 2007; Krzewski et al., 2013), and transmembrane protein 106B (TMEM106B), which inhibits retrograde transport of lysosomes in neuronal dendrites through interaction with the microtubule-associated protein 6 (MAP6) (Schwenk et al., 2014). Finally, the snapin subunit of the BORC and BLOC-1 complexes (Pu et al., 2015) has also been reported to couple lysosomes to dynein, particularly in hippocampal neurons (Cai et al., 2010). This role of snapin is surprising because, as a subunit of BORC, it is involved in coupling of these organelles to kinesin-1 (Pu et al., 2015). Snapin might thus play dual roles in anterograde and retrograde transport as a component of BORC, BLOC-1 or some other complex.

Roles of microtubule motor crosstalk, actin and organelle contacts

The coupling of lysosomes to kinesins and dynein must be tightly regulated to allow for the alternative function of each microtubule motor in anterograde and retrograde transport, respectively. It is conceivable that each of the corresponding GTPases, Arl8 and Rab7, recruits an effector that inactivates the other (e.g. a GAP). Specific phosphoinositides, phosphorylation, ubiquitylation and pH changes are other factors that could control the switch between kinesins and dynein. The mechanisms of a kinesin–dynein switch have been investigated for vesicles carrying amyloid precursor protein (APP) (Fu and Holzbaur, 2013) and brain-derived neurotrophic factor (BDNF) in mammalian neurons (Colin et al., 2008) and early endosomes in filamentous fungi (Schuster et al., 2011), but not yet for late endosomes or lysosomes. In some instances, both kinesins and dynein could act on an organelle at the same time, as is the case for tubular lysosomes in macrophages and dendritic cells, in which the Arl8 and kinesin, and Rab7 and dynein machineries simultaneously pull lysosomes in opposite directions (Mrakovic et al., 2012). In this case, each end of the tubule must behave as a distinct domain capable of recruiting a different motor.

In addition to microtubule motors, the actin cytoskeleton and organelle contacts contribute to the spatial distribution of lysosomes. Interactions with the cortical actin network, in particular, cause transient retention of lysosomes at the cell periphery (Caviston et al., 2011; Encarnação et al., 2016). Lysosomes clustered in the juxtanuclear region upon overexpression of the HOPS subunit VPS18 are also enmeshed in a dense actin network (Poupon et al., 2003), suggesting that there could be a role for the actin cytoskeleton in the central region of the cell as well. In addition, actin filaments and myosin Iα regulate the processivity and directionality of lysosome movement along microtubules (Cordonnier et al., 2001). Contacts with the ER and TGN also modulate the cytoplasmic distribution of lysosomes. To the aforementioned roles of the ER proteins protrudin (Raiborg et al., 2015) and VAP (Rocha et al., 2009) in ER-lysosome contacts, we should add the recently discovered function of the ER-anchored ubiquitin ligase RNF26 (ring finger protein 26) in recruiting the autophagy adaptor p62/sequestosome 1 (SQSTM1) and various ubiquitin-binding organelle adaptors to regulate the positioning of the entire endo-lysosomal system in the perinuclear cloud (Jongsma et al., 2016).

Importance of lysosome movement in cell physiology and pathology

The ability of lysosomes to move within the cytoplasm is crucial to many cellular functions, and perturbations of lysosome movement contribute to the pathogenesis of various diseases. In the next sections, we discuss various physiological and pathological processes in which lysosome movement is involved.

Autophagy

A process that is absolutely dependent on lysosome positioning is autophagy (Yang and Klionsky, 2010). This process starts with the engulfment of cytoplasmic organelles or particles into an autophagosome, which subsequently fuses with a lysosome to generate an autolysosome (Fig. 5). The cytoplasmic materials are thus degraded by lysosomal hydrolases, resulting in the release of breakdown products into the cytosol for subsequent reuse. In addition to the degradation of autophagic substrates, lysosomes serve as a platform for activation of mechanistic target of rapamycin complex 1 (mTORC1), which functions as a negative regulator of autophagy (Noda and Ohsumi, 1998). mTOR is a serine/threonine kinase that controls cell growth and metabolism as part of two multiprotein complexes, mTORC1 and mTORC2 (Zoncu et al., 2011). The discovery that mTORC1 is activated upon translocation from the cytosol to lysosomes has brought new attention to these organelles as a platform for cellular metabolic signaling (Sancak et al., 2008). Nutrient starvation causes mTORC1 to be released from lysosomes, resulting in decreased phosphorylation of two key substrates: the autophagy-initiating UNC51-like kinase 1 (ULK1) and the transcription factor EB (TFEB). Dephosphorylation activates ULK1, promoting the formation of the phagophore, a precursor of the autophagosome (Itakura and Mizushima, 2010), whereas dephosphorylated TFEB translocates from lysosomes to the nucleus to upregulate the expression of autophagy-associated genes (Palmieri et al., 2011; Settembre et al., 2011, 2012; Martina et al., 2012). By co-regulating ULK1 and TFEB, mTORC1 controls autophagy initiation.

Redistribution of lysosomes to the cell periphery mediated by overexpression of the kinesins KIF1Bβ or KIF2, or Arl8b, increases the association of mTORC1 with lysosomes and mTOR activity

Journal of Cell Science (2016) 129, 4329-4339 doi:10.1242/jcs.196287

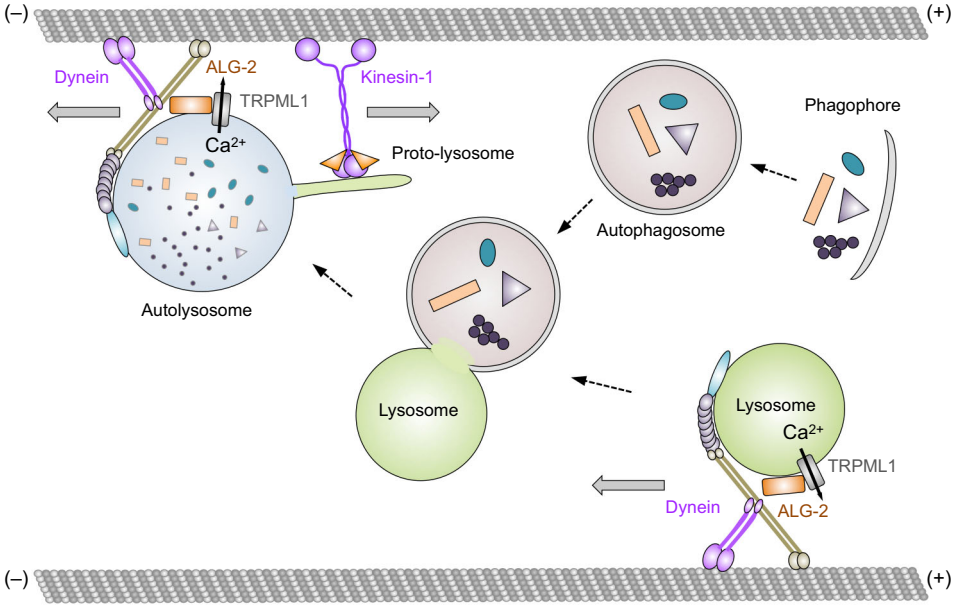

Fig. 5. Lysosome movement in autophagy. Dynein-mediated retrograde transport of lysosomes promotes their fusion with autophagosomes to generate autolysosomes. This transport is sensitive to Ca^{2+} levels, which are sensed through TRPML1 and ALG-2. Autolysosomes also move in the retrograde direction through interaction with dynein. Kinesin-1 pulls tubular proto-lysosomes during autophagic lysosome reformation (ALR).

(Korolchuk et al., 2011). In contrast, juxtanuclear collapse of lysosomes by silencing of KIF2 or Arl8b correlates with reduced mTORC1 activity and increased autophagosome formation (Korolchuk et al., 2011). The increased mTORC1 activity in the periphery could be due to the proximity of lysosomes to the plasma membrane, where mTORC1 activators such as protein kinase B (Akt1) are located. However, knockout of the myrlysin subunit of BORC causes juxtanuclear clustering of lysosomes without changes in mTORC1 activity (Pu et al., 2015). Further studies will thus be needed to ascertain how lysosome positioning influences mTORC1 signaling and autophagy initiation.

Autophagosomes formed in the cell periphery move centripetally along microtubule tracks by means of dynein and subsequently fuse with juxtanuclear lysosomes (Kimura et al., 2008; Korolchuk et al., 2011). Nutrient starvation not only inhibits mTORC1 but also causes juxtanuclear clustering of lysosomes, thereby facilitating autophagosome-lysosome fusion (Korolchuk et al., 2011). Recently, a pathway involving phosphatidylinositol 3,5-bisphosphate [PtdIns(3,5)P_2], the lysosomal Ca^{2+} channel TRPML1, and the Ca^{2+} sensor ALG-2 was shown to participate in this process through regulation of coupling to dynein (Li et al., 2016b). It remains to be determined how Ca^{2+} transients mediated by TRPML1 can promote sustained dynein-dependent motion. Perhaps, Ca^{2+} released by TRPML1 stably binds to the EF hands of ALG-2 in complex with dynein. The lysosome-associated tumor-suppressor protein folliculin has also been found to interact with RILP to promote association with Rab34 on the Golgi and TGN, thus contributing to juxtanuclear clustering of lysosomes in response to nutrient starvation (Starling et al., 2016). Perturbation of these pathways disrupts the juxtanuclear localization of lysosomes, diminishing the probability of autophagosome–lysosome fusion.

Degradation of autophagic substrates is carried out by the lysosomal acid hydrolases. A recent study has shown that juxtanuclear lysosomes are more acidic and have higher cathepsin L activity than peripheral lysosomes (Johnson et al., 2016). This heterogeneity could be due to the stabilization of the V-ATPase subunit V1G1 by RILP (Johnson et al., 2016). The lower pH could explain the higher cathepsin L activity of juxtanuclear lysosomes, and why autophagosomes fuse with juxtanuclear rather then peripheral lysosomes.

The final stage of autophagy is autophagic lysosome reformation (ALR), by which the cellular lysosome pool is restored after autophagy (Yu et al., 2010). During ALR, newly formed tubular lysosomes known as ‘proto-lysosomes’ extend from the autolysosomes and mature to fully functional lysosomes (Fig. 5). A recent study has shown that tubule formation during ALR is driven by the kinesin-1 KIF5B through recruitment to phosphatidylinositol 4,5-bisphosphate [PtdIns(4,5)P_2]-enriched domains on the tubules (Du et al., 2016). The pathway involving PtdIns(3,5)P_2–TRPML1–ALG-2–dynein has also been shown to participate in ALR (Li et al., 2016b).

Immunity

Lysosomes are also capable of killing pathogenic bacteria that enter the cell by way of phagocytosis. To counter this process, bacteria have evolved mechanisms to prevent phagosome maturation and fusion with lysosomes. They do so by injection of various bacterial effectors into the cytosol of the host cell. Some of these effectors modulate the interaction of the bacterial vacuole or lysosomes with the positioning machinery. For example, the *Salmonella enterica* SopD2 effector inactivates Rab7, preventing its interactions with RILP and FYCO1 and thus disrupting transport of lysosomes (D’Costa et al., 2015). Another *Salmonella* effector, SifA, engages SKIP and kinesin-1 to pinch off vesicles from the *Salmonella* vacuole and move them in anterograde direction (Dumont et al., 2010). SifA also forms a complex with SKIP and Rab9A, preventing transport of mannose 6-phosphate receptors (MPRs) from endosomes to the TGN and thus causing lysosomal dysfunction (McGourty et al., 2012). Finally, a third effector, PipB2, interacts with KLC proteins to promote kinesin-1-dependent extension of *Salmonella*-induced filaments as well as movement of lysosomes towards the cell periphery (Knodler and Steele-Mortimer, 2005). The fact the same bacterium encodes multiple effectors to engage the lysosome-positioning machinery emphasizes the importance of this system for infection and immunity.

Digestion of bacteria in lysosomes releases antigenic peptides that bind to MHC-II molecules. In mature dendritic epithelial cells, peptide-loaded MHC-II molecules are transported to the plasma membrane by tubular lysosomes for presentation to CD4-positive T lymphocytes (Chow et al., 2002; Vyas et al., 2007). As mentioned

above, this movement is likely mediated by coupling of lysosomes to kinesin-1 by Arl8, Rab7 and their cognate adaptors (Mrakovic et al., 2012; Saric et al., 2016).

The lysosome-positioning machinery is also crucial for the ability of cytotoxic T lymphocytes (CTLs) and natural killer (NK) cells to kill virally infected or tumor cells. CTLs form immune synapses with their target cells, releasing the contents of lytic granules onto the target cells. Lytic granules contain specific cytotoxic mediators, but also comprise lysosomal luminal and membrane proteins, for which reason they are considered 'lysosome-related organelles' (LROs) (Dell'Angelica et al., 2000) or 'secretory lysosomes' (Blott and Griffiths, 2002). Interestingly, CTLs rely on the same lysosome-positioning machinery as lysosomes to deliver the cytotoxic hit (Fig. 6). After binding to the target cells, lytic granules move towards the MTOC, which in activated CTLs becomes localized under the plasma membrane of the immune synapse (Stinchcombe et al., 2006). This retrograde movement is mediated by the Rab7–RILP–dynein ensemble (Daniele et al., 2011). The Arl8–SKIP–kinesin-1 pathway, by contrast, is required to position the MTOC and lytic granules to the immune synapse in NK cells (Tuli et al., 2013). A Rab27a–Slp3–kinesin-1 complex has also been shown to mediate the terminal movement from the MTOC to the immune synapse (Kurowska et al., 2012). Once under the plasma membrane, lytic granules are transferred from microtubules to filamentous actin by a mechanism involving myosin IIA in NK cells (Andzelm et al., 2007) and the Rab27a–Munc13-4 complex (Munc13-4 is also known as UNC13D) in CTLs (Neeft et al., 2005). Interference with any of these processes reduces the ability of CTLs to kill target cells.

Cancer

Lysosome movement towards the cell periphery is also required for cancer growth, invasion and metastasis. During oncogenic transformation, lysosomes undergo changes in number, morphology, luminal pH, hydrolase content and intracellular distribution (Kroemer and Jäättelä, 2005; Kallunki et al., 2013). A particularly notable change is a shift of the lysosome population from the central to the peripheral cytoplasm (Nishimura et al., 2002, 2003). This redistribution can be induced by changes in the tumor microenvironment, such as acidification (Glunde et al., 2003), or in the expression of genes that regulate lysosome positioning and motility during oncogenic transformation. In this latter regard, various cancer tissues have been shown to exhibit elevated levels of mRNA for KIF5B. Moreover, expression of an active form of the ErbB2 oncogene in a breast cancer cell line increased the amounts of KIF5B and its association with lysosomes (Cardoso et al., 2009). In addition, Rab7 mRNA levels have been found to be downregulated in biopsies of prostate cancer epithelial cells, but not in the surrounding stromal cells or in tissue from benign prostatic hyperplasia (Steffan et al., 2014). This change in Rab7 levels is thought to be pathogenic because peripheral redistribution of lysosomes caused by Rab7 knockdown can be associated with increased cancer cell invasiveness (Steffan et al., 2014). In contrast, depletion of Arl8b prevents lysosome dispersal in response to hepatocyte growth factor, epidermal growth factor and acidic extracellular pH, and diminishes invasive growth of prostate cancer cells (Dykes et al., 2016).

Centrifugal transport is essential for lysosome exocytosis (Rodriguez et al., 1997; Reddy et al., 2001), which results in the release of acid hydrolases into the extracellular space and insertion of late endosomal and/or lysosomal membrane proteins into the plasma membrane. Secreted acid hydrolases are thought to degrade the extracellular matrix, facilitating cell migration and invasion (Mohamed and Sloane, 2006; Dykes et al., 2016). Transport of the transmembrane type 1 matrix metalloproteinase (MT1-MMP, also known as MMP14) from late endosomes to plasma membrane invadopodia also contributes to cancer cell invasiveness and

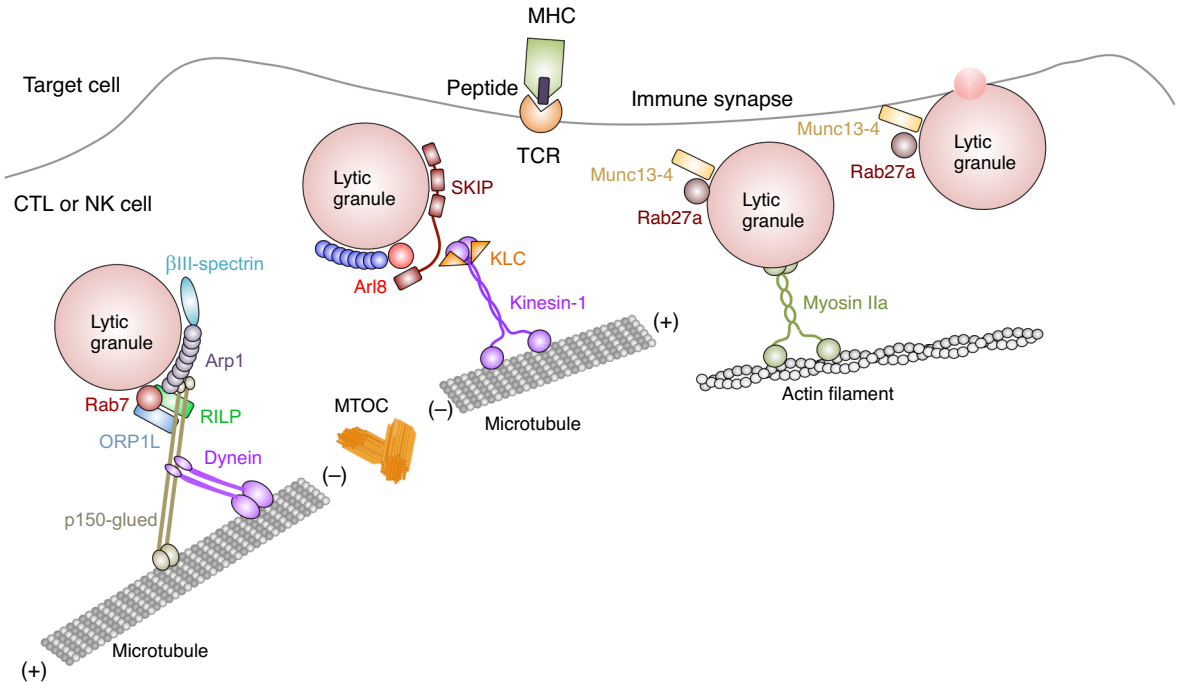

Fig. 6. Lytic granule movement in cytotoxic T lymphocytes and natural killer cells. The scheme integrates findings from both cytotoxic T lymphocytes (CTLs) and natural killer (NK) types, although not all steps might be applicable to both cell types. The MTOC is relocated under the immune synapse. Dynein transports lytic granules from the cell center to the area underlying the immune synapse. Kinesin-1 then moves lytic granules closer to the plasma membrane, where myosin IIa attaches them to actin filaments, and Rab27a and Munc13-4 promote their fusion with the plasma membrane. TCR, T-cell receptor.

metastasis (Monteiro et al., 2013; Macpherson et al., 2014). Furthermore, some integrins that regulate cell adhesion and migration by attachment to the extracellular matrix traffic through late endosomes and lysosomes (Dozynkiewicz et al., 2012), possibly depending on lysosome exocytosis for their delivery to the plasma membrane. Lysosome transport to the cell periphery could also contribute to cell adhesion and migration by regulating the dynamics of focal adhesions. In this regard, Arl8- and kinesin-1-dependent transport of the lysosome-associated ragulator complex has been shown to be essential for removal of IQGAP1 from focal adhesions, enhancing their dynamics (Schiefermeier et al., 2014). The BORC complex also influences cell adhesion and migration, and should therefore be regarded as a possible modulator of cancer cell motility (Pu et al., 2015).

Taken together, these findings emphasize the importance of the centrifugal transport of lysosomes for lysosome exocytosis, extracellular matrix degradation, cell adhesion and migration, all processes that contribute to cancer. In light of these findings, the regulation of lysosome positioning and motility should be considered a potential target for anti-cancer therapeutics (Circu et al., 2016).

Neurological diseases

Although the regulation of lysosome dynamics is important in most cells, it is particularly crucial in neurons because of their extreme asymmetry and the length of axons and dendrites. Indeed, variations or mutations in components of the lysosome-positioning machinery cause various psychiatric and neurological disorders. For example, single nucleotide polymorphisms causing increased expression of the diaskedin (BORCS7) subunit of BORC are a major risk factor for schizophrenia (Duarte et al., 2016; Li et al., 2016a). Additional components of this machinery are mutated in several neurological diseases, including the p150-glued subunit of dynactin in ALS (Munch et al., 2004), Rab7 in Charcot–Marie–Tooth disease type 2B (Verhoeven et al., 2003), KIF1B in Charcot–Marie–Tooth disease type 2A (Zhao et al., 2001), KIF5A in hereditary spastic paraplegia type 10 (Reid et al., 2002) and KIF5C in cortical dysplasia with other brain malformations type 2 (Poirier et al., 2013). Similarly, upregulation of KLC2 is the cause of spastic paraplegia, optic atrophy and neuropathy (SPOAN) syndrome (Melo et al., 2015). Although altered lysosome dynamics could contribute to the pathogenesis of these diseases, the microtubule motors and regulators also mediate transport of additional organelles, such as synaptic vesicle precursors, mitochondria, retrograde transport carriers, and others. Further studies are thus needed to determine the exact contribution of altered lysosome dynamics to the pathogenesis of these diseases.

Some neurological disorders exhibiting altered lysosome positioning are caused by mutations in proteins that are not directly involved in lysosome movement, although they might influence it under physiological and pathological conditions. For example, expression of mutant huntingtin (the causal agent in Huntington's disease) or mutant leucine-rich repeat kinase 2 (LRRK2) (the most common causal agent in familial Parkinson's disease) promote accumulation of lysosomes in the juxtanuclear area of the cell (Caviston et al., 2011; Erie et al., 2015). These effects involve changes in mTORC1, dynein and Rab7 (Caviston et al., 2011; Erie et al., 2015), although the exact mechanisms remain to be elucidated.

Changes in lysosome positioning and motility have also been observed in LSDs, a subset of which affects the central nervous system. In these diseases, mutations in lysosomal proteins lead to the accumulation of storage material in enlarged lysosomal vacuoles. Interestingly, the abnormal lysosomes are often clustered in the juxtanuclear area, as documented for neuronal ceroid lipofuscinosis type 3 (CLN3) (Uusi-Rauva et al., 2012), and mucolipidosis type IV (ML-IV) (Li et al., 2016b). Inhibition of lysosomal proteolysis has also been found to disrupt axonal transport of late endosomes, lysosomes and autolysosomes in neurons, resulting in their accumulation in dystrophic axonal swellings characteristic of Alzheimer's disease (Lee et al., 2011). It is currently unknown, however, how defects in lysosomal hydrolases are translated into changes in lysosome positioning and motility.

Perspectives

From the above discussion, it is clear that lysosome positioning and movement depend on a complex interplay of interactions with microtubule motors, the actin cytoskeleton and other organelles. Although the molecules responsible for these interactions are now beginning to be identified, many more are likely to be uncovered in the future. Outstanding questions to be addressed in future work include: how do lysosomes switch between a static and motile status, and between anterograde and retrograde movement? Why are so many kinesins, kinesin adaptors and dynein adaptors involved in lysosome movement? How are the mechanisms of lysosome transport adapted to polarized cells such as neurons and epithelial cells, which have distinct microtubules in different regions of the cell? How is lysosome movement regulated by nutrient signaling, extracellular pH changes, cellular stresses, infection by intracellular pathogens, oncogenic transformation, and other physiological and pathological conditions? Do specific lipids, such as phosphoinositides, and post-translational modifications, such as phosphorylation and ubiquitylation, participate in the regulation of lysosome movement? How is lysosome transport coordinated with that of related organelles, including endosomes, autophagosomes and the ER? Is there a program for transcriptional control of lysosome positioning? Obtaining answers to these questions will not only shed light on an important physiological process, but also provide clues for therapeutic intervention in lysosomal diseases. We think that the time is now ripe to understand lysosome dynamics in the context of whole cells and organisms, and to exploit this knowledge for technological and medical applications.

Competing interests

The authors declare no competing or financial interests.

Funding

Work in the authors' laboratory is funded by the intramural program of National Institute of Child Health and Human Development (NICHD) (ZIA HD001607). Deposited in PMC for release after 12 months.

References

Aittaleb, M., Chen, P.-J. and Akaaboune, M. (2015). Failure of lysosome clustering and positioning in the juxtanuclear region in cells deficient in rapsyn. *J. Cell Sci.* **128**, 3744-3756.

Al-Bassam, J., Roger, B., Halpain, S. and Milligan, R. A. (2007). Analysis of the weak interactions of ADP-Unc104 and ADP-kinesin with microtubules and their inhibition by MAP2c. *Cell Motil. Cytoskeleton* **64**, 377-389.

Andzelm, M. M., Chen, X., Krzewski, K., Orange, J. S. and Strominger, J. L. (2007). Myosin IIA is required for cytolytic granule exocytosis in human NK cells. *J. Exp. Med.* **204**, 2285-2291.

Baas, P. W., Deitch, J. S., Black, M. M. and Banker, G. A. (1988). Polarity orientation of microtubules in hippocampal neurons: uniformity in the axon and nonuniformity in the dendrite. *Proc. Natl. Acad. Sci. USA* **85**, 8335-8339.

Bagshaw, R. D., Callahan, J. W. and Mahuran, D. J. (2006). The Arf-family protein, Arl8b, is involved in the spatial distribution of lysosomes. *Biochem. Biophys. Res. Commun.* **344**, 1186-1191.

Ballabio, A. (2016). The awesome lysosome. *EMBO Mol. Med.* **8**, 73-76.

Bentley, M., Decker, H., Luisi, J. and Banker, G. (2015). A novel assay reveals preferential binding between Rabs, kinesins, and specific endosomal subpopulations. *J. Cell Biol.* **208**, 273-281.
Blott, E. J. and Griffiths, G. M. (2002). Secretory lysosomes. *Nat. Rev. Mol. Cell Biol.* **3**, 122-131.
Boucrot, E., Henry, T., Borg, J. P., Gorvel, J. P. and Meresse, S. (2005). The intracellular fate of Salmonella depends on the recruitment of kinesin. *Science* **308**, 1174-1178.
Braulke, T. and Bonifacino, J. S. (2009). Sorting of lysosomal proteins. *Biochim. Biophys. Acta* **1793**, 605-614.
Brown, C. L., Maier, K. C., Stauber, T., Ginkel, L. M., Wordeman, L., Vernos, I. and Schroer, T. A. (2005). Kinesin-2 is a motor for late endosomes and lysosomes. *Traffic* **6**, 1114-1124.
Burkhardt, J. K., Echeverri, C. J., Nilsson, T. and Vallee, R. B. (1997). Overexpression of the dynamitin (p50) subunit of the dynactin complex disrupts dynein-dependent maintenance of membrane organelle distribution. *J. Cell Biol.* **139**, 469-484.
Cai, D., McEwen, D. P., Martens, J. R., Meyhofer, E. and Verhey, K. J. (2009). Single molecule imaging reveals differences in microtubule track selection between kinesin motors. *PLoS Biol.* **7**, e1000216.
Cai, Q., Lu, L., Tian, J.-H., Zhu, Y.-B., Qiao, H. and Sheng, Z.-H. (2010). Snapin-regulated late endosomal transport is critical for efficient autophagy-lysosomal function in neurons. *Neuron* **68**, 73-86.
Cantalupo, G., Alifano, P., Roberti, V., Bruni, C. B. and Bucci, C. (2001). Rab-interacting lysosomal protein (RILP): the Rab7 effector required for transport to lysosomes. *EMBO J.* **20**, 683-693.
Cardoso, C. M. P., Groth-Pedersen, L., Høyer-Hansen, M., Kirkegaard, T., Corcelle, E., Andersen, J. S., Jäättelä, M. and Nylandsted, J. (2009). Depletion of kinesin 5B affects lysosomal distribution and stability and induces peri-nuclear accumulation of autophagosomes in cancer cells. *PLoS ONE* **4**, e4424.
Caviston, J. P., Zajac, A. L., Tokito, M. and Holzbaur, E. L. (2011). Huntingtin coordinates the dynein-mediated dynamic positioning of endosomes and lysosomes. *Mol. Biol. Cell* **22**, 478-492.
Chen, Y. and Yu, L. (2013). Autophagic lysosome reformation. *Exp. Cell Res.* **319**, 142-146.
Chow, A., Toomre, D., Garrett, W. and Mellman, I. (2002). Dendritic cell maturation triggers retrograde MHC class II transport from lysosomes to the plasma membrane. *Nature* **418**, 988-994.
Chu, B.-B., Liao, Y.-C., Qi, W., Xie, C., Du, X., Wang, J., Yang, H., Miao, H. H., Li, B.-L. and Song, B.-L. (2015). Cholesterol transport through lysosome-peroxisome membrane contacts. *Cell* **161**, 291-306.
Circu, M. L., Dykes, S. S., Carroll, J., Kelly, K., Galiano, F., Greer, A., Cardelli, J. and El-Osta, H. (2016). A novel high content imaging-based screen identifies the anti-helminthic niclosamide as an inhibitor of lysosome anterograde trafficking and prostate cancer cell invasion. *PLoS ONE* **11**, e0146931.
Colin, E., Zala, D., Liot, G., Rangone, H., Borrell-Pagès, M., Li, X.-J., Saudou, F. and Humbert, S. (2008). Huntingtin phosphorylation acts as a molecular switch for anterograde/retrograde transport in neurons. *EMBO J.* **27**, 2124-2134.
Cordonnier, M.-N., Dauzonne, D., Louvard, D. and Coudrier, E. (2001). Actin filaments and myosin I alpha cooperate with microtubules for the movement of lysosomes. *Mol. Biol. Cell* **12**, 4013-4029.
Daniele, T., Hackmann, Y., Ritter, A. T., Wenham, M., Booth, S., Bossi, G., Schintler, M., Auer-Grumbach, M. and Griffiths, G. M. (2011). A role for Rab7 in the movement of secretory granules in cytotoxic T lymphocytes. *Traffic* **12**, 902-911.
D'costa, V. M., Braun, V., Landekic, M., Shi, R., Proteau, A., McDonald, L., Cygler, M., Grinstein, S. and Brumell, J. H. (2015). Salmonella disrupts host endocytic trafficking by SopD2-mediated inhibition of Rab7. *Cell Rep.* **12**, 1508-1518.
DeBoer, S. R., You, Y., Szodorai, A., Kaminska, A., Pigino, G., Nwabuisi, E., Wang, B., Estrada-Hernandez, T., Kins, S., Brady, S. T. et al. (2008). Conventional kinesin holoenzymes are composed of heavy and light chain homodimers. *Biochemistry* **47**, 4535-4543.
Dehmelt, L. and Halpain, S. (2005). The MAP2/Tau family of microtubule-associated proteins. *Genome Biol.* **6**, 204.
Dell'angelica, E. C., Mullins, C., Caplan, S. and Bonifacino, J. S. (2000). Lysosome-related organelles. *FASEB J.* **14**, 1265-1278.
Dodson, M. W., Zhang, T., Jiang, C., Chen, S. and Guo, M. (2012). Roles of the Drosophila LRRK2 homolog in Rab7-dependent lysosomal positioning. *Hum. Mol. Genet.* **21**, 1350-1363.
Dozynkiewicz, M. A., Jamieson, N. B., MacPherson, I., Grindlay, J., Van Den Berghe, P. V. E., Von Thun, A., Morton, J. P., Gourley, C., Timpson, P., Nixon, C. et al. (2012). Rab25 and CLIC3 collaborate to promote integrin recycling from late endosomes/lysosomes and drive cancer progression. *Dev. Cell* **22**, 131-145.
Du, W., Su, Q. P., Chen, Y., Zhu, Y., Jiang, D., Rong, Y., Zhang, S., Zhang, Y., Ren, H., Zhang, C. et al. (2016). Kinesin 1 drives autolysosome tubulation. *Dev. Cell* **37**, 326-336.
Duarte, R. R., Troakes, C., Nolan, M., Srivastava, D. P., Murray, R. M. and Bray, N. J. (2016). Genome-wide significant schizophrenia risk variation on chromosome 10q24 is associated with altered cis-regulation of BORCS7, AS3MT, and NT5C2 in the human brain. *Am. J. Med. Genet. B Neuropsychiatr. Genet.* **171**, 806-814.
Dumont, A., Boucrot, E., Drevensek, S., Daire, V., Gorvel, J.-P., Poüs, C., Holden, D. W. and Méresse, S. (2010). SKIP, the host target of the Salmonella virulence factor SifA, promotes kinesin-1-dependent vacuolar membrane exchanges. *Traffic* **11**, 899-911.
Dunn, S., Morrison, E. E., Liverpool, T. B., Molina-París, C., Cross, R. A., Alonso, M. C. and Peckham, M. (2008). Differential trafficking of Kif5c on tyrosinated and detyrosinated microtubules in live cells. *J. Cell Sci.* **121**, 1085-1095.
Dykes, S. S., Gray, A. L., Coleman, D. T., Saxena, M., Stephens, C. A., Carroll, J. L., Pruitt, K. and Cardelli, J. A. (2016). The Arf-like GTPase Arl8b is essential for three-dimensional invasive growth of prostate cancer in vitro and xenograft formation and growth in vivo. *Oncotarget* **7**, 31037-31052.
Encarnação, M., Espada, L., Escrevente, C., Mateus, D., Ramalho, J., Michelet, X., Santarino, I., Hsu, V. W., Brenner, M. B., Barral, D. et al. (2016). A Rab3a-dependent complex essential for lysosome positioning and plasma membrane repair. *J. Cell Biol.* **213**, 631-640.
Erie, C., Sacino, M., Houle, L., Lu, M. L. and Wei, J. (2015). Altered lysosomal positioning affects lysosomal functions in a cellular model of Huntington's disease. *Eur. J. Neurosci.* **42**, 1941-1951.
Falcon-Perez, J. M., Starcevic, M., Gautam, R. and Dell'angelica, E. C. (2002). BLOC-1, a novel complex containing the pallidin and muted proteins involved in the biogenesis of melanosomes and platelet-dense granules. *J. Biol. Chem.* **277**, 28191-28199.
Fraldi, A., Klein, A. D., Medina, D. L. and Settembre, C. (2016). Brain disorders due to lysosomal dysfunction. *Annu. Rev. Neurosci.* **39**, 277-295.
Frasa, M. A., Maximiano, F. C., Smolarczyk, K., Francis, R. E., Betson, M. E., Lozano, E., Goldenring, J., Seabra, M. C., Rak, A., Ahmadian, M. R. et al. (2010). Armus is a Rac1 effector that inactivates Rab7 and regulates E-cadherin degradation. *Curr. Biol.* **20**, 198-208.
Fu, M.-M. and Holzbaur, E. L. F. (2013). JIP1 regulates the directionality of APP axonal transport by coordinating kinesin and dynein motors. *J. Cell Biol.* **202**, 495-508.
Ganley, I. G. and Pfeffer, S. R. (2006). Cholesterol accumulation sequesters Rab9 and disrupts late endosome function in NPC1-deficient cells. *J. Biol. Chem.* **281**, 17890-17899.
Ganley, I. G., Carroll, K., Bittova, L. and Pfeffer, S. (2004). Rab9 GTPase regulates late endosome size and requires effector interaction for its stability. *Mol. Biol. Cell* **15**, 5420-5430.
Glunde, K., Guggino, S. E., Solaiyappan, M., Pathak, A. P., Ichikawa, Y. and Bhujwalla, Z. M. (2003). Extracellular acidification alters lysosomal trafficking in human breast cancer cells. *Neoplasia* **5**, 533-545.
Guardia, C. M., Farías, G. G., Jia, R., Pu, J. and Bonifacino, J. S. (2016). BORC functions upstream of kinesins 1 and 3 to coordinate regional movement of lysosomes along different microtubule tracks. *Cell Reports*, in press.
Hammond, J. W., Cai, D. and Verhey, K. J. (2008). Tubulin modifications and their cellular functions. *Curr. Opin. Cell Biol.* **20**, 71-76.
Harada, A., Takei, Y., Kanai, Y., Tanaka, Y., Nonaka, S. and Hirokawa, N. (1998). Golgi vesiculation and lysosome dispersion in cells lacking cytoplasmic dynein. *J. Cell Biol.* **141**, 51-59.
Heuser, J. (1989). Changes in lysosome shape and distribution correlated with changes in cytoplasmic pH. *J. Cell Biol.* **108**, 855-864.
Hirokawa, N. and Noda, Y. (2008). Intracellular transport and kinesin superfamily proteins, kifs: structure, function, and dynamics. *Physiol. Rev.* **88**, 1089-1118.
Hofmann, I. and Munro, S. (2006). An N-terminally acetylated Arf-like GTPase is localised to lysosomes and affects their motility. *J. Cell Sci.* **119**, 1494-1503.
Hollenbeck, P. J. and Swanson, J. A. (1990). Radial extension of macrophage tubular lysosomes supported by kinesin. *Nature* **346**, 864-866.
Huynh, K. K., Eskelinen, E. L., Scott, C. C., Malevanets, A., Saftig, P. and Grinstein, S. (2007). LAMP proteins are required for fusion of lysosomes with phagosomes. *EMBO J.* **26**, 313-324.
Ikegami, K., Heier, R. L., Taruishi, M., Takagi, H., Mukai, M., Shimma, S., Taira, S., Hatanaka, K., Morone, N., Yao, I. et al. (2007). Loss of α-tubulin polyglutamylation in ROSA22 mice is associated with abnormal targeting of KIF1A and modulated synaptic function. *Proc. Natl Acad. Sci. USA* **104**, 3213-3218.
Ishikawa, T. (2012). Structural biology of cytoplasmic and axonemal dyneins. *J. Struct. Biol.* **179**, 229-234.
Itakura, E. and Mizushima, N. (2010). Characterization of autophagosome formation site by a hierarchical analysis of mammalian Atg proteins. *Autophagy* **6**, 764-776.
Jackson, L. K., Nawabi, P., Hentea, C., Roark, E. A. and Haldar, K. (2008). The Salmonella virulence protein SifA is a G protein antagonist. *Proc. Natl. Acad. Sci. USA* **105**, 14141-14146.
Johansson, M., Rocha, N., Zwart, W., Jordens, I., Janssen, L., Kuijl, C., Olkkonen, V. M. and Neefjes, J. (2007). Activation of endosomal dynein motors by stepwise assembly of Rab7-RILP-p150Glued, ORP1L, and the receptor betalII spectrin. *J. Cell Biol.* **176**, 459-471.

Johnson, D. E., Ostrowski, P., Jaumouillé, V. and Grinstein, S. (2016). The position of lysosomes within the cell determines their luminal pH. *J. Cell Biol.* **212**, 677-692.
Jongsma, M. L. M., Berlin, I., Wijdeven, R. H. M., Janssen, L., Janssen, G. M. C., Garstka, M. A., Janssen, H., Mensink, M., Van Veelen, P. A., Spaapen, R. M. et al. (2016). An ER-associated pathway defines endosomal architecture for controlled cargo transport. *Cell* **166**, 152-166.
Jordens, I., Fernandez-Borja, M., Marsman, M., Dusseljee, S., Janssen, L., Calafat, J., Janssen, H., Wubbolts, R. and Neefjes, J. (2001). The Rab7 effector protein RILP controls lysosomal transport by inducing the recruitment of dynein-dynactin motors. *Curr. Biol.* **11**, 1680-1685.
Kallunki, T., Olsen, O. D. and Jäättelä, M. (2013). Cancer-associated lysosomal changes: friends or foes? *Oncogene* **32**, 1995-2004.
Kevenaar, J. T., Bianchi, S., Van Spronsen, M., Olieric, N., Lipka, J., Frias, C. P., Mikhaylova, M., Harterink, M., Keijzer, N., Wulf, P. S. et al. (2016). Kinesin-binding protein controls microtubule dynamics and cargo trafficking by regulating kinesin motor activity. *Curr. Biol.* **26**, 849-861.
Kimura, S., Noda, T. and Yoshimori, T. (2008). Dynein-dependent movement of autophagosomes mediates efficient encounters with lysosomes. *Cell Struct. Funct.* **33**, 109-122.
Knodler, L. A. and Steele-Mortimer, O. (2005). The Salmonella effector PipB2 affects late endosome/lysosome distribution to mediate Sif extension. *Mol. Biol. Cell* **16**, 4108-4123.
Ko, D. C., Gordon, M. D., Jin, J. Y. and Scott, M. P. (2001). Dynamic movements of organelles containing Niemann-Pick C1 protein: NPC1 involvement in late endocytic events. *Mol. Biol. Cell* **12**, 601-614.
Konishi, Y. and Setou, M. (2009). Tubulin tyrosination navigates the kinesin-1 motor domain to axons. *Nat. Neurosci.* **12**, 559-567.
Korolchuk, V. I., Saiki, S., Lichtenberg, M., Siddiqi, F. H., Roberts, E. A., Imarisio, S., Jahreiss, L., Sarkar, S., Futter, M., Menzies, F. M. et al. (2011). Lysosomal positioning coordinates cellular nutrient responses. *Nat. Cell Biol.* **13**, 453-460.
Kroemer, G. and Jäättelä, M. (2005). Lysosomes and autophagy in cell death control. *Nat. Rev. Cancer* **5**, 886-897.
Krzewski, K., Gil-Krzewska, A., Nguyen, V., Peruzzi, G. and Coligan, J. E. (2013). LAMP1/CD107a is required for efficient perforin delivery to lytic granules and NK-cell cytotoxicity. *Blood* **121**, 4672-4683.
Kurowska, M., Goudin, N., Nehme, N. T., Court, M., Garin, J., Fischer, A., de Saint Basile, G. and Menasche, G. (2012). Terminal transport of lytic granules to the immune synapse is mediated by the kinesin-1/Slp3/Rab27a complex. *Blood* **119**, 3879-3889.
Lebrand, C., Corti, M., Goodson, H., Cosson, P., Cavalli, V., Mayran, N., Faure, J. and Gruenberg, J. (2002). Late endosome motility depends on lipids via the small GTPase Rab7. *EMBO J.* **21**, 1289-1300.
Lee, S., Sato, Y. and Nixon, R. A. (2011). Lysosomal proteolysis inhibition selectively disrupts axonal transport of degradative organelles and causes an Alzheimer's-like axonal dystrophy. *J. Neurosci.* **31**, 7817-7830.
Li, M., Jaffe, A. E., Straub, R. E., Tao, R., Shin, J. H., Wang, Y., Chen, Q., Li, C., Jia, Y., Ohi, K. et al. (2016a). A human-specific AS3MT isoform and BORCS7 are molecular risk factors in the 10q24.32 schizophrenia-associated locus. *Nat. Med.* **22**, 649-656.
Li, X., Rydzewski, N., Hider, A., Zhang, X., Yang, J., Wang, W., Gao, Q., Cheng, X. and Xu, H. (2016b). A molecular mechanism to regulate lysosome motility for lysosome positioning and tubulation. *Nat. Cell Biol.* **18**, 404-417.
Lin, S. X. and Collins, C. A. (1992). Immunolocalization of cytoplasmic dynein to lysosomes in cultured cells. *J. Cell Sci.* **101**, 125-137.
Lipka, J., Kapitein, L. C., Jaworski, J. and Hoogenraad, C. C. (2016). Microtubule-binding protein doublecortin-like kinase 1 (DCLK1) guides kinesin-3-mediated cargo transport to dendrites. *EMBO J.* **35**, 302-318.
Loubéry, S., Wilhelm, C., Hurbain, I., Neveu, S., Louvard, D. and Coudrier, E. (2008). Different microtubule motors move early and late endocytic compartments. *Traffic* **9**, 492-509.
Macpherson, I. R., Rainero, E., Mitchell, L. E., Van Den Berghe, P. V. E., Speirs, C., Dozynkiewicz, M. A., Chaudhary, S., Kalna, G., Edwards, J., Timpson, P. et al. (2014). CLIC3 controls recycling of late endosomal MT1-MMP and dictates invasion and metastasis in breast cancer. *J. Cell Sci.* **127**, 3893-3901.
Martina, J. A., Chen, Y., Gucek, M. and Puertollano, R. (2012). MTORC1 functions as a transcriptional regulator of autophagy by preventing nuclear transport of TFEB. *Autophagy* **8**, 903-914.
Marx, A., Müller, J., Mandelkow, E.-M., Hoenger, A. and Mandelkow, E. (2005). Interaction of kinesin motors, microtubules, and MAPs. *J. Muscle Res. Cell Motil.* **27**, 125-137.
Matsushita, M., Tanaka, S., Nakamura, N., Inoue, H. and Kanazawa, H. (2004). A novel kinesin-like protein, KIF1Bβ3 is involved in the movement of lysosomes to the cell periphery in non-neuronal cells. *Traffic* **5**, 140-151.
Matsuzaki, F., Shirane, M., Matsumoto, M. and Nakayama, K. I. (2011). Protrudin serves as an adaptor molecule that connects KIF5 and its cargoes in vesicular transport during process formation. *Mol. Biol. Cell* **22**, 4602-4620.
Matteoni, R. and Kreis, T. E. (1987). Translocation and clustering of endosomes and lysosomes depends on microtubules. *J. Cell Biol.* **105**, 1253-1265.
McGourty, K., Thurston, T. L., Matthews, S. A., Pinaud, L., Mota, L. J. and Holden, D. W. (2012). Salmonella inhibits retrograde trafficking of mannose-6-phosphate receptors and lysosome function. *Science* **338**, 963-967.
Melo, U. S., Macedo-Souza, L. I., Figueiredo, T., Muotri, A. R., Gleeson, J. G., Coux, G., Armas, P., Calcaterra, N. B., Kitajima, J. P., Amorim, S. et al. (2015). Overexpression of KLC2 due to a homozygous deletion in the non-coding region causes SPOAN syndrome. *Hum. Mol. Genet.* **24**, 6877-6885.
Mizuno, K., Kitamura, A. and Sasaki, T. (2003). Rabring7, a novel Rab7 target protein with a RING finger motif. *Mol. Biol. Cell* **14**, 3741-3752.
Mohamed, M. M. and Sloane, B. F. (2006). Cysteine cathepsins: multifunctional enzymes in cancer. *Nat. Rev. Cancer* **6**, 764-775.
Monteiro, P., Rossé, C., Castro-Castro, A., Irondelle, M., Lagoutte, E., Paul-Gilloteaux, P., Desnos, C., Formstecher, E., Darchen, F., Perrais, D. et al. (2013). Endosomal WASH and exocyst complexes control exocytosis of MT1-MMP at invadopodia. *J. Cell Biol.* **203**, 1063-1079.
Moriyama, K. and Bonifacino, J. S. (2002). Pallidin is a component of a multi-protein complex involved in the biogenesis of lysosome-related organelles. *Traffic* **3**, 666-677.
Mrakovic, A., Kay, J. G., Furuya, W., Brumell, J. H. and Botelho, R. J. (2012). Rab7 and Arl8 GTPases are necessary for lysosome tubulation in macrophages. *Traffic* **13**, 1667-1679.
Mukhopadhyay, R. (2016). Lively lysosomes. *ASBMB Today* **15**, 7.
Munch, C., Sedlmeier, R., Meyer, T., Homberg, V., Sperfeld, A. D., Kurt, A., Prudlo, J., Peraus, G., Hanemann, C. O., Stumm, G. et al. (2004). Point mutations of the p150 subunit of dynactin (DCTN1) gene in ALS. *Neurology* **63**, 724-726.
Nakata, T. and Hirokawa, N. (1995). Point mutation of adenosine triphosphate-binding motif generated rigor kinesin that selectively blocks anterograde lysosome membrane transport. *J. Cell Biol.* **131**, 1039-1053.
Nakata, T., Niwa, S., Okada, Y., Perez, F. and Hirokawa, N. (2011). Preferential binding of a kinesin-1 motor to GTP-tubulin-rich microtubules underlies polarized vesicle transport. *J. Cell Biol.* **194**, 245-255.
Neeft, M., Wieffer, M., De Jong, A. S., Negroiu, G., Metz, C. H., Van Loon, A., Griffith, J., Krijgsveld, J., Wulffraat, N., Koch, H. et al. (2005). Munc13-4 is an effector of rab27a and controls secretion of lysosomes in hematopoietic cells. *Mol. Biol. Cell* **16**, 731-741.
Nishimura, Y., Itoh, K., Yoshioka, K., Ikeda, K. and Himeno, M. (2002). A role for small GTPase RhoA in regulating intracellular membrane traffic of lysosomes in invasive rat hepatoma cells. *Histochem. J.* **34**, 189-213.
Nishimura, Y., Itoh, K., Yoshioka, K., Tokuda, K. and Himeno, M. (2003). Overexpression of ROCK in human breast cancer cells: evidence that ROCK activity mediates intracellular membrane traffic of lysosomes. *Pathol. Oncol. Res.* **9**, 83-95.
Noda, T. and Ohsumi, Y. (1998). Tor, a phosphatidylinositol kinase homologue, controls autophagy in yeast. *J. Biol. Chem.* **273**, 3963-3966.
Nordmann, M., Cabrera, M., Perz, A., Bröcker, C., Ostrowicz, C., Engelbrecht-Vandré, S. and Ungermann, C. (2010). The Mon1-Ccz1 complex is the GEF of the late endosomal Rab7 homolog Ypt7. *Curr. Biol.* **20**, 1654-1659.
Nottingham, R. M., Pusapati, G. V., Ganley, I. G., Barr, F. A., Lambright, D. G. and Pfeffer, S. R. (2012). RUTBC2 protein, a Rab9A effector and GTPase-activating protein for Rab36. *J. Biol. Chem.* **287**, 22740-22748.
Palmieri, M., Impey, S., Kang, H., di Ronza, A., Pelz, C., Sardiello, M. and Ballabio, A. (2011). Characterization of the CLEAR network reveals an integrated control of cellular clearance pathways. *Hum. Mol. Genet.* **20**, 3852-3866.
Parenti, G., Andria, G. and Ballabio, A. (2015). Lysosomal storage diseases: from pathophysiology to therapy. *Annu. Rev. Med.* **66**, 471-486.
Parton, R. G., Dotti, C. G., Bacallao, R., Kurtz, I., Simons, K. and Prydz, K. (1991). pH-induced microtubule-dependent redistribution of late endosomes in neuronal and epithelial cells. *J. Cell Biol.* **113**, 261-274.
Paschal, B. M. and Vallee, R. B. (1987). Retrograde transport by the microtubule-associated protein MAP 1C. *Nature* **330**, 181-183.
Peralta, E. R., Martin, B. C. and Edinger, A. L. (2010). Differential effects of TBC1D15 and mammalian Vps39 on Rab7 activation state, lysosomal morphology, and growth factor dependence. *J. Biol. Chem.* **285**, 16814-16821.
Poirier, K., Lebrun, N., Broix, L., Tian, G., Saillour, Y., Boscheron, C., Parrini, E., Valence, S., Pierre, B. S., Oger, M. et al. (2013). Mutations in TUBG1, DYNC1H1, KIF5C and KIF2A cause malformations of cortical development and microcephaly. *Nat. Genet.* **45**, 639-647.
Poupon, V., Stewart, A., Gray, S. R., Piper, R. C. and Luzio, J. P. (2003). The role of mVps18p in clustering, fusion, and intracellular localization of late endocytic organelles. *Mol. Biol. Cell* **14**, 4015-4027.
Progida, C., Malerod, L., Stuffers, S., Brech, A., Bucci, C. and Stenmark, H. (2007). RILP is required for the proper morphology and function of late endosomes. *J. Cell Sci.* **120**, 3729-3737.
Pu, J., Schindler, C., Jia, R., Jarnik, M., Backlund, P. and Bonifacino, J. S. (2015). BORC, a multisubunit complex that regulates lysosome positioning. *Dev. Cell* **33**, 176-188.
Raiborg, C., Wenzel, E. M., Pedersen, N. M., Olsvik, H., Schink, K. O., Schultz, S. W., Vietri, M., Nisi, V., Bucci, C., Brech, A. et al. (2015). Repeated ER–

endosome contacts promote endosome translocation and neurite outgrowth. *Nature* **520**, 234-238.
Raiborg, C., Wenzel, E. M., Pedersen, N. M. and Stenmark, H. (2016). ER–endosome contact sites in endosome positioning and protrusion outgrowth. *Biochem. Soc. Trans.* **44**, 441-446.
Reddy, A., Caler, E. V. and Andrews, N. W. (2001). Plasma membrane repair is mediated by Ca(2+)-regulated exocytosis of lysosomes. *Cell* **106**, 157-169.
Reed, N. A., Cai, D., Blasius, T. L., Jih, G. T., Meyhofer, E., Gaertig, J. and Verhey, K. J. (2006). Microtubule acetylation promotes kinesin-1 binding and transport. *Curr. Biol.* **16**, 2166-2172.
Reid, E., Kloos, M., Ashley-Koch, A., Hughes, L., Bevan, S., Svenson, I. K., Graham, F. L., Gaskell, P. C., Dearlove, A., Pericak-Vance, M. A. et al. (2002). A kinesin heavy chain (KIF5A) mutation in hereditary spastic paraplegia (SPG10). *Am. J. Hum. Genet.* **71**, 1189-1194.
Rocha, N., Kuijl, C., van der Kant, R., Janssen, L., Houben, D., Janssen, H., Zwart, W. and Neefjes, J. (2009). Cholesterol sensor ORP1L contacts the ER protein VAP to control Rab7-RILP-p150 Glued and late endosome positioning. *J. Cell Biol.* **185**, 1209-1225.
Rodriguez, A., Webster, P., Ortego, J. and Andrews, N. W. (1997). Lysosomes behave as Ca2+-regulated exocytic vesicles in fibroblasts and epithelial cells. *J. Cell Biol.* **137**, 93-104.
Rosa-Ferreira, C. and Munro, S. (2011). Arl8 and SKIP act together to link lysosomes to kinesin-1. *Dev. Cell* **21**, 1171-1178.
Saftig, P. and Klumperman, J. (2009). Lysosome biogenesis and lysosomal membrane proteins: trafficking meets function. *Nat. Rev. Mol. Cell Biol.* **10**, 623-635.
Sancak, Y., Peterson, T. R., Shaul, Y. D., Lindquist, R. A., Thoreen, C. C., Bar-Peled, L. and Sabatini, D. M. (2008). The Rag GTPases bind raptor and mediate amino acid signaling to mTORC1. *Science* **320**, 1496-1501.
Santama, N., Krijnse-Locker, J., Griffiths, G., Noda, Y., Hirokawa, N. and Dotti, C. G. (1998). KIF2beta, a new kinesin superfamily protein in non-neuronal cells, is associated with lysosomes and may be implicated in their centrifugal translocation. *EMBO J.* **17**, 5855-5867.
Saric, A., Hipolito, V. E. B., Kay, J. G., Canton, J., Antonescu, C. N. and Botelho, R. J. (2016). mTOR controls lysosome tubulation and antigen presentation in macrophages and dendritic cells. *Mol. Biol. Cell* **27**, 321-333.
Schiefermeier, N., Scheffler, J. M., De Araujo, M. E., Stasyk, T., Yordanov, T., Ebner, H. L., Offterdinger, M., Munck, S., Hess, M. W., Wickstrom, S. A. et al. (2014). The late endosomal p14-MP1 (LAMTOR2/3) complex regulates focal adhesion dynamics during cell migration. *J. Cell Biol.* **205**, 525-540.
Schuster, M., Lipowsky, R., Assmann, M.-A., Lenz, P. and Steinberg, G. (2011). Transient binding of dynein controls bidirectional long-range motility of early endosomes. *Proc. Natl. Acad. Sci. USA* **108**, 3618-3623.
Schwenk, B. M., Lang, C. M., Hogl, S., Tahirovic, S., Orozco, D., Rentzsch, K., Lichtenthaler, S. F., Hoogenraad, C. C., Capell, A., Haass, C. et al. (2014). The FTLD risk factor TMEM106B and MAP6 control dendritic trafficking of lysosomes. *EMBO J.* **33**, 450-467.
Settembre, C., Di Malta, C., Polito, V. A., Garcia Arencibia, M., Vetrini, F., Erdin, S., Erdin, S. U., Huynh, T., Medina, D., Colella, P. et al. (2011). TFEB links autophagy to lysosomal biogenesis. *Science* **332**, 1429-1433.
Settembre, C., Zoncu, R., Medina, D. L., Vetrini, F., Erdin, S., Erdin, S., Huynh, T., Ferron, M., Karsenty, G., Vellard, M. C. et al. (2012). A lysosome-to-nucleus signalling mechanism senses and regulates the lysosome via mTOR and TFEB. *EMBO J.* **31**, 1095-1108.
Starcevic, M. and Dell'angelica, E. C. (2004). Identification of snapin and three novel proteins (BLOS1, BLOS2, and BLOS3/reduced pigmentation) as subunits of biogenesis of lysosome-related organelles complex-1 (BLOC-1). *J. Biol. Chem.* **279**, 28393-28401.
Starling, G. P., Yip, Y. Y., Sanger, A., Morton, P. E., Eden, E. R. and Dodding, M. P. (2016). Folliculin directs the formation of a Rab34-RILP complex to control the nutrient-dependent dynamic distribution of lysosomes. *EMBO Rep.* **17**, 823-841.
Steffan, J. J., Dykes, S. S., Coleman, D. T., Adams, L. K., Rogers, D., Carroll, J. L., Williams, B. J. and Cardelli, J. A. (2014). Supporting a role for the GTPase Rab7 in prostate cancer progression. *PLoS One* **9**, e87882.
Stinchcombe, J. C., Majorovits, E., Bossi, G., Fuller, S. and Griffiths, G. M. (2006). Centrosome polarization delivers secretory granules to the immunological synapse. *Nature* **443**, 462-465.
Sung, H.-H., Telley, I. A., Papadaki, P., Ephrussi, A., Surrey, T. and Rørth, P. (2008). Drosophila ensconsin promotes productive recruitment of kinesin-1 to microtubules. *Dev. Cell* **15**, 866-876.
Tanaka, Y., Kanai, Y., Okada, Y., Nonaka, S., Takeda, S., Harada, A. and Hirokawa, N. (1998). Targeted disruption of mouse conventional kinesin heavy chain kif5B, results in abnormal perinuclear clustering of mitochondria. *Cell* **93**, 1147-1158.
Tuli, A., Thiery, J., James, A. M., Michelet, X., Sharma, M., Garg, S., Sanborn, K. B., Orange, J. S., Lieberman, J. and Brenner, M. B. (2013). Arf-like GTPase Arl8b regulates lytic granule polarization and natural killer cell-mediated cytotoxicity. *Mol. Biol. Cell* **24**, 3721-3735.
Uusi-Rauva, K., Kyttälä, A., van der Kant, R., Vesa, J., Tanhuanpää, K., Neefjes, J., Olkkonen, V. M. and Jalanko, A. (2012). Neuronal ceroid lipofuscinosis protein CLN3 interacts with motor proteins and modifies location of late endosomal compartments. *Cell. Mol. Life Sci.* **69**, 2075-2089.
Verhoeven, K., De Jonghe, P., Coen, K., Verpoorten, N., Auer-Grumbach, M., Kwon, J. M., FitzPatrick, D., Schmedding, E., De Vriendt, E., Jacobs, A. et al. (2003). Mutations in the small GTP-ase late endosomal protein RAB7 cause Charcot-Marie-Tooth type 2B neuropathy. *Am. J. Hum. Genet.* **72**, 722-727.
Vyas, J. M., Kim, Y.-M., Artavanis-Tsakonas, K., Love, J. C., Van der Veen, A. G. and Ploegh, H. L. (2007). Tubulation of class II MHC compartments is microtubule dependent and involves multiple endolysosomal membrane proteins in primary dendritic cells. *J. Immunol.* **178**, 7199-7210.
Wang, T. and Hong, W. (2002). Interorganellar regulation of lysosome positioning by the Golgi apparatus through Rab34 interaction with Rab-interacting lysosomal protein. *Mol. Biol. Cell* **13**, 4317-4332.
Wurmser, A. E., Sato, T. K. and Emr, S. D. (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**, 551-562.
Yang, Z. and Klionsky, D. J. (2010). Mammalian autophagy: core molecular machinery and signaling regulation. *Curr. Opin. Cell Biol.* **22**, 124-131.
Yau, K. W., Schatzle, P., Tortosa, E., Pages, S., Holtmaat, A., Kapitein, L. C. and Hoogenraad, C. C. (2016). Dendrites in vitro and in vivo contain microtubules of opposite polarity and axon formation correlates with uniform plus-end-out microtubule orientation. *J. Neurosci.* **36**, 1071-1085.
Yu, L., McPhee, C. K., Zheng, L., Mardones, G. A., Rong, Y., Peng, J., Mi, N., Zhao, Y., Liu, Z., Wan, F. et al. (2010). Termination of autophagy and reformation of lysosomes regulated by mTOR. *Nature* **465**, 942-946.
Yu, L., Wu, W. K., Gu, C., Zhong, D., Zhao, X., Kong, Y., Lin, Q., Chan, M. T., Zhou, Z. and Liu, S. (2016). Obatoclax impairs lysosomal function to block autophagy in cisplatin-sensitive and -resistant esophageal cancer cells. *Oncotarget* **7**, 14693-14707.
Zaarur, N., Meriin, A. B., Bejarano, E., Xu, X., Gabai, V. L., Cuervo, A. M. and Sherman, M. Y. (2014). Proteasome failure promotes positioning of lysosomes around the aggresome via local block of microtubule-dependent transport. *Mol. Cell. Biol.* **34**, 1336-1348.
Zhang, X.-M., Walsh, B., Mitchell, C. A. and Rowe, T. (2005). TBC domain family, member 15 is a novel mammalian Rab GTPase-activating protein with substrate preference for Rab7. *Biochem. Biophys. Res. Commun.* **335**, 154-161.
Zhang, Z., Wang, S., Shen, T., Chen, J. and Ding, J. (2014). Crystal structure of the Rab9A-RUTBC2 RBD complex reveals the molecular basis for the binding specificity of Rab9A with RUTBC2. *Structure* **22**, 1408-1420.
Zhao, C., Takita, J., Tanaka, Y., Setou, M., Nakagawa, T., Takeda, S., Yang, H. W., Terada, S., Nakata, T., Takei, Y. et al. (2001). Charcot-Marie-Tooth disease type 2A caused by mutation in a microtubule motor KIF1Bbeta. *Cell* **105**, 587-597.
Zoncu, R., Bar-Peled, L., Efeyan, A., Wang, S., Sancak, Y. and Sabatini, D. M. (2011). mTORC1 senses lysosomal amino acids through an inside-out mechanism that requires the vacuolar H(+)-ATPase. *Science* **334**, 678-683.