Received: 23 July 2020 | Revised: 4 December 2020 | Accepted: 28 December 2020

DOI: 10.1002/jcb.29889

REVIEW

Journal of Cellular Biochemistry WILEY

Lipid droplets and autophagy—links and regulations from yeast to humans

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Funding information
Deutsche Forschungsgemeinschaft, Grant/Award Number: SFB 1177

Abstract

Recent advances in the yeast *Saccharomyces cerevisiae* and higher eukaryotes have been increasingly connecting lipid droplet (LD) dynamics to the regulation of autophagy. In this review we will discuss implications that connect LD *de novo* synthesis and LD mobilization to autophagy and how autophagy is regulated by these mechanisms. Elucidating these connections might pose a chance to further understand autophagy induction and membrane biogenesis for the growing autophagosome under different conditions. Increasing our understanding of these mechanisms might provide a chance to understand several conditions that might be related to LD dysregulation and, possibly, as a consequence of this, dysregulation of autophagy.

KEYWORDS
autophagy, lipid droplets, lipolysis, lipophagy, mammals, yeast

1 | INTRODUCTION

Macroautophagy, hereafter referred to as autophagy, is a cellular process involved in catabolism of cellular molecules, organelles, and protein aggregates and is conserved across species. In most cellular systems it is thought of as cytoprotective and can be induced as response to several conditions, such as starvation or other cellular stresses, to provide essential nutrients or macromolecules for the cell.[1,2]

Several autophagy related genes (ATGs) have been first described in *S. cerevisiae* to play a crucial role in orchestrating autophagy and have since been described across species.[3,4] The formation of the autophagosome can be divided into several steps, of which each requires recruitment of ATG-complexes. In yeast, cup-shaped membraneous structures, called phagophores, are initially synthesized at the phagophore assembly site (PAS), a process that in the mammalian cell can happen at different sites. After formation, the phagophore-membrane is elongated and forms a spherical structure, which will seal to form the autophagosome. The autophagosome eventually fuses with the vacuole (yeast)/lysosome (mammals) and is, together with its cargo, degraded via hydrolytic activity.[5,6]

Here, we will concentrate on discussing new insight into the connection of autophagy to lipid droplets (LDs). LDs are structures for lipid storage that are conserved across species and consist of a core of triacylglycerols (TAGs) and steryl esters (SEs) surrounded by a monolayer of phospholipids that is decorated with several proteins (Figure 1).[7] They are formed in-between the two membrane leaflets of the endoplasmic reticulum (ER) and, after maturation, bud off but remain functionally connected to the ER.[8,9]

For a long time, LDs have been viewed as structures solely for the purpose of lipid storage in cells that contribute to the cellular energy metabolism. During high-energy availability, LDs are synthesized and, when energy levels fall, they are being broken down to provide nutrients for the cell. However, a growing number of studies have implicated other roles of LDs such as protection against lipotoxicity and oxidative stress.[10]

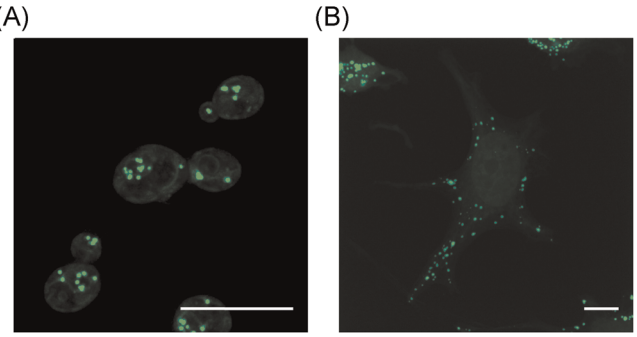

FIGURE 1 Visualization of lipid droplets (LDs) in S. cerevisiae and a HeLa cell. (A) LDs in wild-type yeast cells (BY4741) stained with BODIPY493/503, scale bar = 10 μm. (B) A LDs in a HeLa cell, stained with BODIPY493/503, scale bar = 10 μm

Additionally, studies in the yeast *S. cerevisiae* have shown that LD-dynamics play a role in regulating autophagy, which is inhibited after disruption of neutral lipid synthesis.[11] Furthermore, a mobilization of activated fatty acids (FAs) from LDs has been shown to promote autophagy in several models.[11–14] However, the exact mechanisms that connect the metabolism of lipids of the LDs (hereafter referred to as LD-metabolism) to autophagy are only partly understood. Recent findings suggest that LDs might be a source of lipids for the growing phagophore and that this might be the reason for the more and more apparent connections between LD-dynamics and autophagy. Interestingly, it has recently been found in yeast that the long chain fatty acyl-CoA synthetase Faa1 localizes directly to the growing autophagosome which points towards a direct mechanism for the biogenesis of the autophagosomal membrane at the expanding autophagosome.[15]

Here, we will briefly review and discuss insights into and possible connections between autophagy and LD mobilization, considering recent findings and proposals of how LD dynamics might have an impact on autophagy.

2 | LDS AS REGULATORS OF AUTOPHAGY

2.1 | LD-synthesis and its role in autophagy

As mentioned above, LDs are synthesized between the two membranes of the ER. This process involves several enzymes that ultimately produce the TAGs and SEs for lipid storage (Figure 2A,A′). In this section, we will discuss the implications of TAG and SE synthesis in regulating autophagy.

2.1.1 | TAG-biogenesis and autophagy

To synthesize TAGs, activity of the yeast enzyme lipin Pah1, an Mg^{2+}-dependent phosphatidate phosphatase, is required to convert phosphatidic acid (PA) to diacylglycerol (DAG). Pah1 is dephosphorylated by the ER-membrane associated protein complex Nem1-Sop7 whereupon it associates to perinuclear ER membranes, bringing it to the proximity of its substrate PA.[16,17] Upon DAG synthesis via Pah1, DAG is converted to TAG by two diacylglycerol-transferases Dga1 and Lro1. Dga1 binds to the ER-membrane as well as the phospholipid monolayer of the forming LD, which is thought to tether the forming LD to the ER membrane.[8] Dga1 is active during stationary phase growth, synthesizing TAGs using different acyl-CoA species.[18] Lro1 is bound to the ER-membrane only and, in contrast to Dga1, uses the phospholipids PE or PC, to synthesize TAG in an acyl-CoA-independent manner. It is active during exponential growth.[9]

In mammalian cells, TAG-synthesis is achieved in a similar manner and is, in human cells, mediated by the diacylglycerol-acyltransferases DGAT1 and DGAT2, homologs of yeast Dga1 and Lro1, respectively.[10]

The above described mechanisms have been implicated to play a role in the regulation of autophagy. A study in yeast showed that Nem1-Spo7 and, consequently, Pha1 activity is required for the induction of autophagy. Upon deletion of either Nem1-Spo7 or Pah1, autophagy-induction, via inactivation of TORC1, was shown to be compromised.[19] Similarly, another study showed that a double deletion of Dga1 and Lro1 partially reduces autophagic flux.[20]

To date, the exact molecular mechanisms connecting proteins that are involved in TAG synthesis to autophagy induction are still unclear. However, it has been shown that a deletion of Dga1 an Lro1 causes failure of ATG8 lipidation.[11] As lipidation of Atg8 (LC3 family and GABARAP family human cells) is known to be essential for the recruitment of autophagy-related genes to the growing autophagosome and, thereby, for autophagy progression itself,[21] it is convincing that failure to lipidate Atg8 in Dga1 and Lro1 deletion strains causes inhibition of autophagy. Furthermore, it was foundis that a deletion of Dga1 and Lro1 in yeast causes changes in the phospholipid composition in the cell that are likely to impact ER-homeostasis and, thereby, autophagic flux.[20]

TAG synthesis and expression of DGAT1 and DGAT2 have been demonstrated to be increased under nitrogen starvation conditions in mammalian cells. It was also revealed, that an inhibition of DGAT1 in mammalian cells has little to no effect on autophagic flux under nitrogen starvation conditions and might rather serve as a

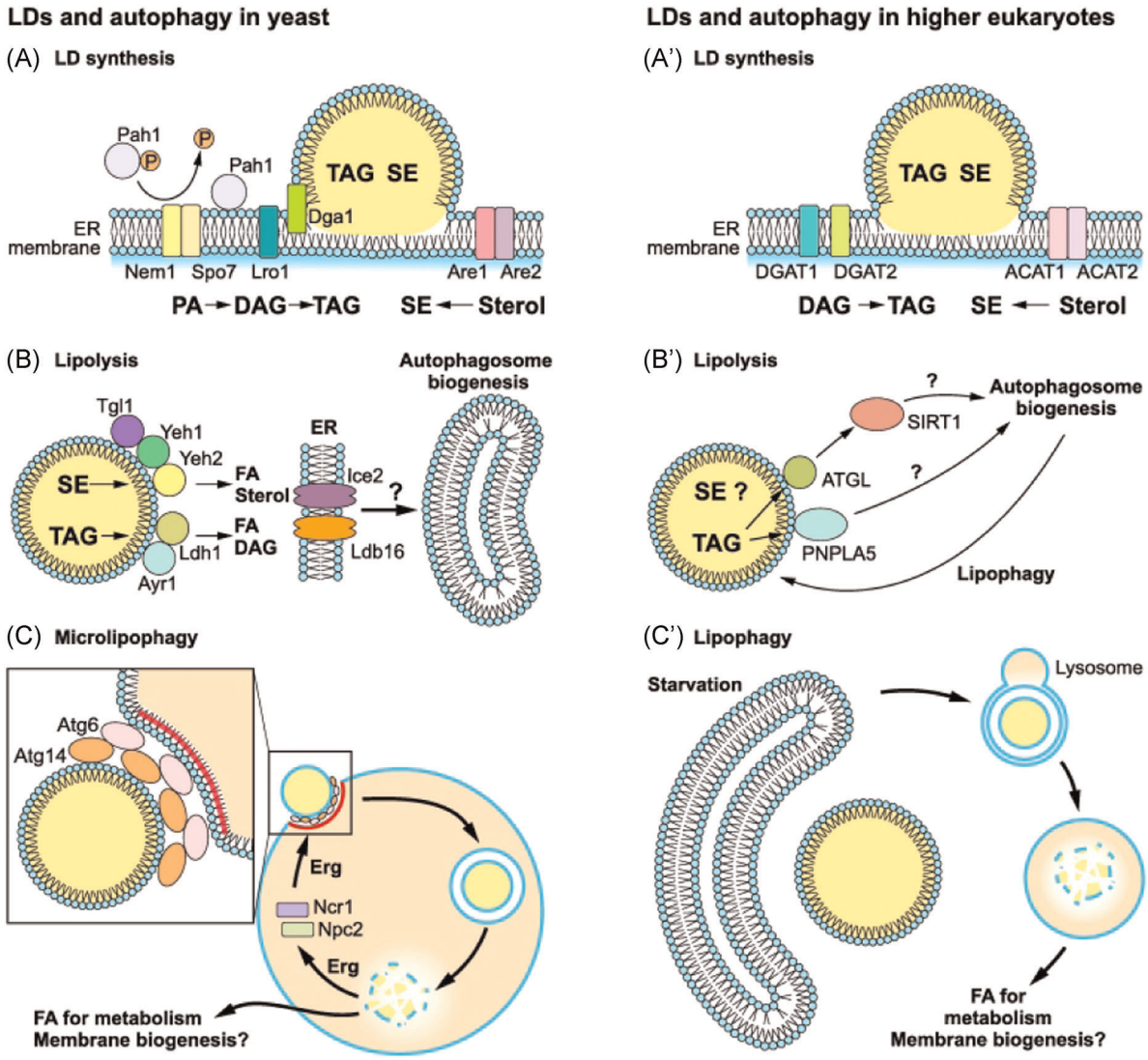

FIGURE 2 Factors in LD dynamics that are connected to autophagy in yeast and higher eukaryotes. (A) LD-synthesis in yeast. TAG synthesis is regulated via Nem1/Spo7 mediated Pah1 dephosphorylation whereupon Pah1 localizes to the ER-membrane and catalyses the reaction from PA to DAG. Lro1 and Dga1 mediate the reaction from DAG to TAG. When deleted, all factors have a negative impact on autophagy-initiation. SE-synthesis is mediated by Are1 and Are2. In Are1 and Are2 deletion strains, autophagy initiation and autophagic flux is inhibited. When all factors (Lro1, Dga1, Are1, and Are2) of TAG and SE synthesis are deleted the cell lacks LDs and autophagy is inhibited. (A′) LD synthesis in higher eukaryotes. TAG synthesis is mediated by DGAT1 and DGAT2. In a DGAT1 KO mouse, it was found that lipophagy is less sufficient and that lysosomes are less well acidified. SE synthesis is mediated by ACAT1 and ACAT2, however, little is known about SE synthesis in higher eukaryotes and in connection to autophagy. (B) Lipolysis in yeast. The lipases Tgl1, Yeh1, and Yeh2 break down SE to free FAs and sterols and the lipases Ldh1 and Ayr1 break down TAG to free FAs and DAG. A deletion of these lipases has been shown to be essential for autophagy. Ice2 and Ldb16 maintain LD-ER contact sites and are required for FA channeling for ER membrane synthesis. A deletion of these factors reduces autophagy; however, it remains unclear whether they also channel FAs for the synthesis of autophagosomal membranes. (B′) Lipolysis in higher eukaryotes. In liver cells, TAG is mobilized by ATGL and, together with SIRT1, induces lipophagy. Little is known about the exact mechanism. Similarly, the lipase PNPLA5 is required for autophagy. (C) Microlipophagy in yeast releases FAs for metabolism. LDs are directly incorporated into the vacuole. This process requires factors of the core autophagy machinery, such as Atg6 and Atg14. The incorporation sites are sterol-rich microdomains (red membranes) that are maintained by Ncr1 and Npc2. It is still unclear whether the free FAs may contribute to autophagosome biogenesis. (C′) Lipophagy in higher eukaryotes. Small LDs are enveloped in LC3-positive membranes and are broken down via lipophagy, which releases free FAs for metabolism. The exact mechanisms are still unclear. ATGL, adipose triglyceride lipase; DAG, diacylglycerol; ER, endoplasmic reticulum; FA, fatty acid; KO, knockout; LD, lipid droplet; PA, phosphatidic acid; SE, steryl ester; SIRT1, sirtuin 1; TAG, triacylglycerol

mechanism to sequester toxic acylcarnitine in LDs that occur during autophagic breakdown of, for example, mitochondria.[22]

However, a more recent study, analyzing intestinal cells of Dgat KO mice, found that lipophagy, a selective form of autophagy implied in LD-breakdown (discussed below), is less sufficient and that lysosomes are less sufficiently acidified.[23]

2.1.2 | SE-biogenesis and autophagy

Together with TAGs, SEs are stored in LDs. SE synthesis is mediated by the two acyl Coenzyme A:sterol acyltransferases Are1 and Are2 in yeast. These enzymes esterify unsaturated acyl-CoA species to form SEs and are, like the TAG-synthesis machinery, located to the ER-membrane. In human cells, the homologs of Are1 and Are2, ACAT1 and ACAT2, function in a similar manner. However, to date, little is known about SE synthesis and its connection to autophagy in higher eukaryotes. It has been shown that a deletion of Are1 and Are2 in yeast, similarly to a deletion of Dga1 and Lro1, reduces the autophagic flux. When all four enzymes are deleted, the cell is completely devoid of LDs and autophagy is inhibited.[11,20] In microglia, an inhibition of ACAT1 increases autophagy and clearance of Alzheimer-associated Aβ1-42 aggregates. Later, this was also shown for an inhibition of ACAT1 in a mouse model for Alzheimer's disease (AD).[24,25] It was suggested, that ACAT1 might regulate overall cholesterol levels in the cells which is detrimental in models for AD.[26,27] Cholesterol (or egosterol in yeast) has also been implied in vacuolar fusion processes. In yeast models, the presence of ergosterol in membrane rafts can promote membrane fusion and is necessary for vacuolar ATPase activity.[28]

Taken together, the above described findings might suggest a vital role of LD biogenesis in enabling autophagic flux. There is increasing evidence that correct regulation of lipid homeostasis seems to be important for several cellular processes, including autophagy, ER-membrane homeostasis and vacuolar fusion processes. There is also some evidence that suggests LDs being involved in protecting the cell against lipotoxicity. How these findings connect to each other remains to be further investigated. Given that, for example, autophagy plays a vital role in several human diseases such as, for example, the majority of neurodegenerative diseases or cancer, describing the mechanisms that connect LD biogenesis and autophagy and how aberrant or increased LD synthesis might regulate autophagy has great potential in increasing our knowledge about disease formation.

3 | LD-MOBILIZATION AS LIPID SOURCE FOR THE AUTOPHAGOSOME

LD-mobilization has, similarly to LD-synthesis, been connected to autophagy in several ways. One major pathway of LD-breakdown has been suggested to be so called lipophagy with the purpose of supplying free FAs for the cell's energy metabolism (Figure 2C,C′). However, other studies have implied a role of lipolysis contributing to the regulation of autophagy itself (Figure 2B,B′). In this section, we discuss the current knowledge about the mechanisms of lipophagy and lipolysis in connection to autophagy.

3.1 | Lipophagy

In mammals, it has been implied that LDs are broken down by autophagy. Studies have shown that, upon nutrient starvation, LDs associate with autophagic components and that TAG components of LDs increase.[29] Further, the autophagosomal membrane marker protein LC3, a member of the Atg8 family, was detected in LDs, suggesting that autophagy plays a role in LD regulation.[29,30] Other studies found evidence for lipophagy in several tissues such as, for example, agouti-related peptide neurons,[31] macrophage foam cells,[32] and adipocytes. In adipocytes nutrient restriction upregulates the transcription factor forkhead homeobox type protein O1 which subsequently enhances expression of the lysosomal acid lipase (Lipa). Together with the observation that, under these conditions, LDs have been found to co-localize with lysosomes, it was suggested that Lipa might mediate LD-degradation by lipophagy.[33] However, the exact mechanisms of lipophagy in mammalian cells still remain to be elucidated. Furthermore, LDs have been implied in contributing to the formation of the autophagosomal membrane (see below) and yeast cells that lack LDs also show a lack of autophagic flux (see above). Thus, it remains unclear how lipophagy on the one hand, and a contribution of LDs to the formation of autophagosomes on the other hand are exactly regulated.

Studies in yeast have implied a degradation of LDs via a process called microautophagy. It has been shown that, at the shift of exponential- to stationary-phase growth conditions, LDs are directly incorporated into the vacuole (lysosome) and play a role in maintaining microdomains within the vacuole that, in turn, promote microlipophagy. These microdomains are likely sterol-rich domains and are organized by the action of the Niemann-Pick type C (NPC) proteins Ncr1 and Npc2.[34]

Furthermore, parts of the core autophagy machinery and functioning macroautophagy are required for micro-lipophagy.[35] To date, it is still unclear whether micro-lipophagy, as it has been described in yeast, also exists in mammalian cells.

This implies a strong interconnection between different forms of autophagy and LD break-down and might suggest highly diverse functions for proteins involved in autophagy as well as of proteins regulating LD homeostasis.

3.2 | Lipolysis

Several lipases, that degrade TAGs and SEs, are localized to LDs. These lipases are known to contribute to LD homeostasis and are thought to provide building blocks for cellular membranes, including autophagosomal membranes, as well as to aid cellular adaptation to different energy requirements. In this section we will discuss the role of several lipases and their implication in autophagy.

In yeast, lipases, that degrade TAG to DAG and FAs, have been described to be Tgl3, Tgl4, Tgl5, Ayr1, and Ldh1, which are localized to LDs. Tgl3, Tgl4, and Tgl5 are major lipases in *S. cerevisiae* and catalyze the hydrolysis of TAGs to DAGs and free FAs.[36–39] While it has been shown that Tgl3, Tgl4, and Tgl5 are necessary for TAG mobilization for β-oxidation and metabolism, they have not yet been identified as essential for autophagy induction (Table 1). In contrast to this, the activity of the TAG lipases Ayr1 and Ldh1 appears to be essential for bulk autophagy in yeast. Interestingly, deletion strains of these enzymes cannot be rescued by addition of FAs, suggesting a specialized role for Ayr1 and Ldh1 in autophagy.[11] Next to its TAG lipase activity, Ayr1 is a 1-acyl dihydroxyacetone phosphate reductase (ADR) (Table 1), which is involved in the synthesis of PA, a (low abundancy) phospholipid in cellular membranes.[40,41] Interestingly, PA has been implied in autophagosome formation and fusion as well as an activator of the mammalian target of rapamycin.[41–43] Thus, one could hypothesize that Ayr1's implication in autophagy might be due to its additional function as ADR. However, it is still not entirely clear what exactly the mechanisms, that aid autophagy, are.

In mammals, neutral lipases, possibly together with autophagy, have been described to aid FA mobilization for β-oxidation under starvation conditions.[44] However, another study has shown that the mammalian neutral lipase, phospholipase domain-containing protein 5, is required for autophagy initiation and localizes to LDs. It has been proposed that this mechanism contributes to the formation of the autophagosomal membrane (Table 2).[12] Another lipase that has been suggested to be involved in the promotion of autophagy/lipophagy is the adipose triglyceride lipase (ATGL). It has been shown that this promotion of autophagy and lipophagy is induced by ATGL-mediated activation of sirtuin 1 (Table 2).[45] These findings suggest a more complex role of neutral lipases in mammalian cells, possibly integrating the regulation of cellular metabolism with autophagy or lipophagy induction.

SE-mobilization in yeast is mediated by the SE-hydrolases Yeh1, Yeh2, and Tgl1.[46–48] Similar to the Ayr1-Ldh1 deletion strains, deletions of Yeh1, Yeh2, and Tgl1 are essential for bulk autophagy in yeast.[11] Furthermore, efficient channeling of the DAGs and FAs, that are released by the action of the yeast lipases Ayr1, Ldh1, Yeh1, Yeh2, and Tgl1, is required for initiation of autophagy. A deletion of the enzymes Ice2 and Ldb16, that maintain LD-ER contact sites, have also been demonstrated to reduce autophagy after starvation.[11] It has further been shown that Ice2 and Ldb16 regulate LD size and are required for free FA and DAG channeling for ER-membrane synthesis.[49,50] Furthermore, a recent study uncovered that Ice2 negatively regulates Pah1, thereby opposing the Nem7-Spo7 complex. This activity may shift the lipid metabolism towards membrane biogenesis of the ER.[51] This might also be true for the autophagosomal membrane.

Taken together, these findings suggest that appropriate mobilization and channeling of DAGs and free FAs might be crucial for functional autophagy. Mobilization of free FAs and DAGs are suggested to contribute to the formation of phospholipids for the formation of membranes, including autophagosomal membranes.

4 | AUTOPHAGY AS POTENTIAL REGULATOR OF LD DYNAMICS

To date, a lot of research has been focussing on how LD dynamics regulate autophagy, however, very little is known about whether autophagy itself might be regulating LD-dynamics. The most obvious example of this idea is LDs being degraded by lipophagy. On top of that, another possibility would be a regulation of LD dynamics via targeted autophagosomal degradation of proteins involved in, for example LD synthesis or tethering of LDs. This might provide a possibility to regulate LD dynamics and, as a consequence of that, might regulate autophagy itself.

Some factors that play a role in autophagy have already been connected to play additional roles in LD dynamics and lipid distribution throughout the cell.

TABLE 1 Yeast proteins involved in LD dynamics that are connected to autophagy (after: Li et. al.[52]; Sphilka et al.[20]; Athenstaedt & Daum[40])

Protein	Mechanism	Localization	Influence on autophagy
TAG biogenesis			
Nem1-Spo7 complex	Complex with Spo7, dephosphorylation and activation of Pah1	ER-membrane	Activity required for autophagy induction
Pah1	DAG synthesis	Dephosphorylated Pah1 localizes to ER-membrane	Activity required for autophagy induction
Dga1	TAG-synthase, catalyses formation of TAG from DAG	ER-membrane	Double deletion inhibits autophagy and shows a lack of Atg8-lipidation
Lro1	TAG-synthase, catalyses formation of TAG from PE or PC	ER-membrane	
Ice2	LD-ER contact site, channeling of FAs, thought to determine LD number, negative regulation of Pah1	ER-Membrane at LD-ER contact sites	Autophagy is inhibited in deletion strain
Ldb16	LD-ER contact site, channeling of FAs, though to determine LD-size	ER-Membrane, at LD-ER contact sites	Autophagy is inhibited in deletion strain
SE biogenesis			
Are1	SE synthesis from unsaturated acyl-CoA	ER-Membrane	Double deletion reduces autophagic flux
Are2	SE synthesis from unsaturated acyl-CoA	ER-Membrane	
Lipolysis of TAGs			
Tgl3	TAG lipase	LDs	Triple deletion has little effect on autophagy
Tgl4	TAG lipase	LDs	
Tgl5	TAG lipase	LDs	
Ayr1	TAG lipase and 1-acyl DHAP reductase activity	LDs, ER-membrane and outer mitochondrial membrane	Autophagy is inhibited in deletion strain
Ldh1	TAG lipase	LDs	Autophagy is inhibited in deletion strain
Lipolysis of SEs			
Yeh1	SE hydrolase	LDs	Autophagy is inhibited in deletion strain
Yeh2	SE hydrolase, paralog of Yeh1	LDs	Autophagy is inhibited in deletion strain
Tgl1	SE hydrolase	LDs	Autophagy is inhibited in deletion strain

Abbreviations: DAG, diacylglycerol; ER, endoplasmic reticulum; FA, fatty acid; LD, lipid droplet; PA, phosphatidic acid; PC, phosphatidylcholine; TAG, triacylglycerol.

4.1 | Atg2

A knock down of ATG2A and ATG2B (the mammalian homologs of the yeast Atg2), but not a knock-down of ATG5, in cells causes an accumulation of enlarged LDs and inhibits autophagic flux.[53] Furthermore, yeast Atg2 and mammalian ATG2A have been shown to tether the growing end of the autophagosomal membrane to the ER and to mediate phospholipid transport for the growing phagophore. It has been suggested the structure of Atg2 and ATG2A contain a hydrophobic cavity that facilitates the transport of phospholipids from the ER to the phagohore.[54–56] Interestingly, ATG2A also localizes, together with ATG14L, to LDs where is has been suggested to play a role in regulating LD number and size.[57] While it is still unclear how ATG2-mediated LD regulation might connect to autophagy, these results suggest an involvement of an Atg protein in the regulation of LDs and this might lead to the suggestion, that other Atg proteins could also help the regulation of LDs in the cell. Whether this is achieved through lipophagy or other processes remains to be investigated.

TABLE 2 Mammalian proteins involved in LD dynamics that are connected to autophagy (Dupont et al.,[12] Hung & Buhman,[23] Shibuya et al.,[24] and Kahn et al.[45])

Protein	Mechanism	Localization	Influence on autophagy
TAG biogenesis			
DGAT1	DAG-acyltransferase, catalyses formation of TAG from DAG and FA	ER-membrane	KO mice show less sufficient lipophagy and autophagosome acidification
DGAT2	DAG-acyltransferase, catalyses formation of TAG from DAG and FA	ER-membrane	-
SE biogenesis			
ACAT1	Sterol-acyltransferase, catalyses formation of SEs	ER-membrane	Inhibition in murine microglia stimulates autophagy
ACAT2	Sterol-acyltransferase, catalyses formation of SEs	ER-membrane	-
Lipolysis			
PNPLA5	TAG-lipase	LDs	Required for autophagy initiation
ATGL	TAG-lipase	LDs	ATGL-mediated activation of SIRT1promotes lipophagy/autophagy in hepatocytes

Abbreviations: ATGL, adipose triglyceride lipase; DAG, diacylglycerol; ER, endoplasmic reticulum; FA, fatty acid; KO, knockout; LD, lipid droplet; PNPLA5, phospholipase domain-containing protein 5; SIRT1, sirtuin 1; TAG, triacylglycerol.

4.2 | RAB18

In mammalian cells, the small GTPase RAB18 contributes to the regulation of autophagy via stimulation by the RAB guanine nucleotide exchange factors RAB3GAP1 and RAB3GAP2. A deletion of RAB18 inhibits autophagic activity and decreases LD numbers.[58–60] To regulate LDs, activated RAB18 binds to LDs and bind to the ER via the ER-associated NAG-RINT1-ZW10 complex and its associated SANREs.[60] Furthermore, the autophagy-modulating function of RAB18 is likely caused by the release of LD-derived FAs.[61] Interstingly, a loss of function mutation of RAB18 is associated with Warburg micro syndrome,[62] a (neuro-) developmental disorder. Loss of RAB18 in a mouse model has been suggested to cause accumulation of neurofilament and microtubule proteins.[63]

While there is little known about the specific pathways that connect regulation of autophagy to LD dynamics, the current knowledge might point towards a more interconnected regulatory machinery of both, LD dynamics and autophagy. Thus, it might be possible that more factors that are known to regulate autophagy will be found to regulate LD dynamics as well.

5 | LDS AND AUTOPHAGY IN NEURODEGENERATION

LDs have been connected to a number of human diseases. The most obvious examples might be obesity and nonalcoholic fatty liver disease.[10] Another group of disease connected to LD dysregulation is cancer. It was found that LDs play a role during cancer formation and progression.[64,65]

Interestingly, some studies have emerged recently, that have connected LD accumulation in microglia of the ageing mouse brain and accumulation of LDs in Drosophila glia cells promote neurodegeneration.[66,67] Recently, an accumulation of LDs has been identified in the ageing brain in vivo and in a mouse model for frontotemporal dementia, suggesting a more direct connection between LD accumulation and neurodegeneration.[68] To date, LD accumulation in microglia has been mainly connected to inflammation and stress.[69] However, it might be possible that, like in other models, LD accumulation, cause by e.g. dysregulation, might have an impact on autophagy and might thus play a role during or at the onset of neurodegeneration. Interestingly, in Parkinson's disease (PD) models, LD dysregulation has recently been suggested as potentially being the actual neurotoxic process that might be caused by alpha synuclein (aS) accumulation.[70] Even though PD as “lipidopathy” is still being discussed, it has previously been shown that aS aggregates from on LD membranes, potentially inhibiting lipolysis.[71] Work in a yeast model demonstrated that expressing WT and A53T aS causes accumulation of LDs.[72] More recent work suggests that an inhibition of the stearoyl-CoA-desaturase, involved in oleic acid synthesis, protects against aS toxicity in yeast.[73] Similarly, in AD, an accumulation of LDs in mouse models has been described.[74] On top of that, other neurodegenerative disorders have been described to be associated with LD dysregulation (reviewed in[75]:).

Traditionally, Neurodegeneration has been connected to dysregulation of the autophagy-lysosomal pathway.[76] In AD, it was shown, that autophagy-related compartments were enriched in neuronal cells and were involved in Aβ-formation.[77] Later it was shown, that a number of other neurodegenerative diseases, such as PD, Huntington's disease, or Amyotrophic lateral sclerosis can be caused by a dysfunction of the autophagy–lysosomal pathway.[78] Interestingly, it has been suggested that lysosomes become increasingly dysfunctional in ageing microglial cells.[79]

While a lot of research has focussed on autophagy or LD dynamics in neurodegeneration, how these connect to cell homeostasis, aging, and neurodegeneration still need to be described in more detail. Further understanding why LDs accumulate in the ageing brain and how this connects to neurodegeneration and autophagy might help understand these diseases and better understanding will pose a chance for new treatment approaches.

6 | CONCLUDING REMARKS

Even though there have been a lot of advances and new findings in the field, the question of how autophagy exactly connects to LD dynamics and where the membranes of the growing autophagosomes are derived from are still unclear. A lot of data suggests several mechanisms that might exist in parallel or might be regulated by yet unknown mechanisms. It is also possible that these mechanisms are connected but that the connecting factors are, to date, not described. This could be the case for ATG2A and ATG14L, for example, of which the localization and its importance in autophagosome formation suggests that it might be a tether of several organelles, such as the ER, LDs, and the nascent autophagosome to ensure lipid trafficking across those membranes. It is entirely possible that these lipids might be derived from FAs that have been mobilized from LDs.

The idea that LDs might serve as lipid source for the nascent autophagosomal membrane is supported by several studies and LD homeostasis might be a major factor for the regulation of autophagy simply by changing the availability of lipids that are needed for the biogenesis of the autophagosome. Interestingly, recent findings have suggested a localization of Faa1 directly to the growing autophagosome, which might suggest a more direct mechanism for membrane biogenesis at the autophagosomal membrane itself.[15]

Another question, that remains unclear, is whether different autophagy pathways might source membranes via different mechanisms. It has for example been described that, in yeast, a form of microlipophagy exists but, interestingly, in mammals, lipophagy via microautophagy has not been described. Taken together, LD dynamics increasingly prove to be major players in regulating autophagy. As insufficient autophagy has been implied in several diseases, such as several neurodegenerative diseases and cancer, elucidating the mechanisms of LD dynamics in connection to autophagy might give more insight into disease mechanisms.

ACKNOWLEDGMENTS

The authors would like to thank Dr. Paraskevi Kritsiligkou, Dr. Andreas Kern, Daniel Schmitt and Fazilet Bekbulat for their help and excellent feedback on the manuscript. The work of The Autophagy Lab Mainz is funded by the Deutsche Forschungsgemeinschaft (DFG, German Research Foundation)—Project-ID 259130777-SFB 1177 and the Peter-Beate-Heller-Stiftung.

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How to cite this article: Schepers J, Behl C. Lipid droplets and autophagy—links and regulations from yeast to humans. *J Cell Biochem*. 2021;122:602–611. https://doi.org/10.1002/jcb.29889